Debrecen University
Centre for Agricultural Sciences
Faculty of Agriculture

3rd International Plant Protection Symposium (3rdIPPS)at Debrecen University

(8th Trans-Tisza Plant Protection Forum)

From ideas to implementation

Challenge and Practice of Plant Protection in the beginning of the 21st century

15-16 October, 2003
Debrecen, Hungary

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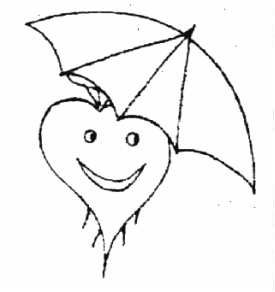

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LECTURES OF PLENARY SESSION

EUROPE-WIDE PHEROMONE STUDIES ON CLICK BEETLES (COLEOPTERA: ELATERIDAE)

Mikós Tóth[1] – Lorenzo Furlan[2] – István Szarukán[3] – István Ujváry[4] – Venyamin G. Yatsynin[5]

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Wireworms, the larvae of click beetles (Coleoptera, Elateridae), rank among the most important soil-dwelling agricultural pests worldwide. In most countries insecticides are applied to soil on a schedule, without actual risk assessment of wireworm damage, mostly because of the clumsiness and labor-intensiveness of conventional methods of population sampling and density estimation for these pests. In Italy, for example, of the total area treated with soil insecticides, only a small percentage is actually in economic danger of wireworm attack (Furlan, 1989, Furlan et al., 2002).

Trapping the adults could assist in making long-term forecast decisions (more than a year) on the need for soil insecticide treatments in the area in question. Similar to other groups of insect pests, sex pheromone baited traps would be ideal monitoring tools (Furlan et al., 1997).

Click beetles form a well distinct, more or less uniform group within Coleoptera, both by taxonomy, and by other life habits.

Evidence for the existence of long-range sex pheromones within this taxonomic group have been demonstrated for a number of species from different continents (Borg-Karlson et al., 1988, Ivaschenko and Adamenko, 1980, Kamm et al., 1983, Yatsynin et al., 1980). Most studies on this subject deal with Euroasian spp. and originate from scientists from the former Soviet Union (Kudryavtsev et al., 1993, Siirde et al., 1993, Yatsynin et al., 1996).

We have recently developed pheromone baits and traps for catching males of all important pest click beetles in Central and Western Europe. Traps and baits were optimized in tests conducted at several sites mainly in Hungary, Italy and Switzerland. This optimization was also performed with species for which previous information on pheromone composition of populations from Russia had been available, as the occurrence of different pheromonal strains or ecotypes within the same species, depending on geographical occurrence, is not uncommon. Finally, the most effective pheromone

combinations for each species were tested in a Europe-wide comparative effort. In the present paper we summarize results of these studies.

Materials and Methods

Connected to the genitals of female click beetles there is a bulbous gland-like structure, which emits its content into the ovipositor. According to general opinion this organ stores the pheromone produced by the insect (Oleschenko et al., 1976, Ivaschenko and Adamenko, 1980). Female sex pheromone gland extracts from reared or collected individuals of all species were prepared as described by carefully piercing the pheromone gland by a fine glass capillary and collecting the liquid inside into the capillary (Oleschenko et al., 1976, Ivaschenko and Adamenko, 1980). The samples obtained were dissolved in hexane, and were analyzed by capillary gas chromatography – mass spectrometry. The identified structures were synthetized and their biological activity was studied in electrophysiological and field trapping tests. Experiments aimed at the optimization of bait composition, and of trap design were conducted first of all in Italy, Hungary and Switzerland. Traps baited with the optimized baits of each species were sent to many countries of Europe, where local cooperators parallely conducted field trapping tests.

Results and Discussion

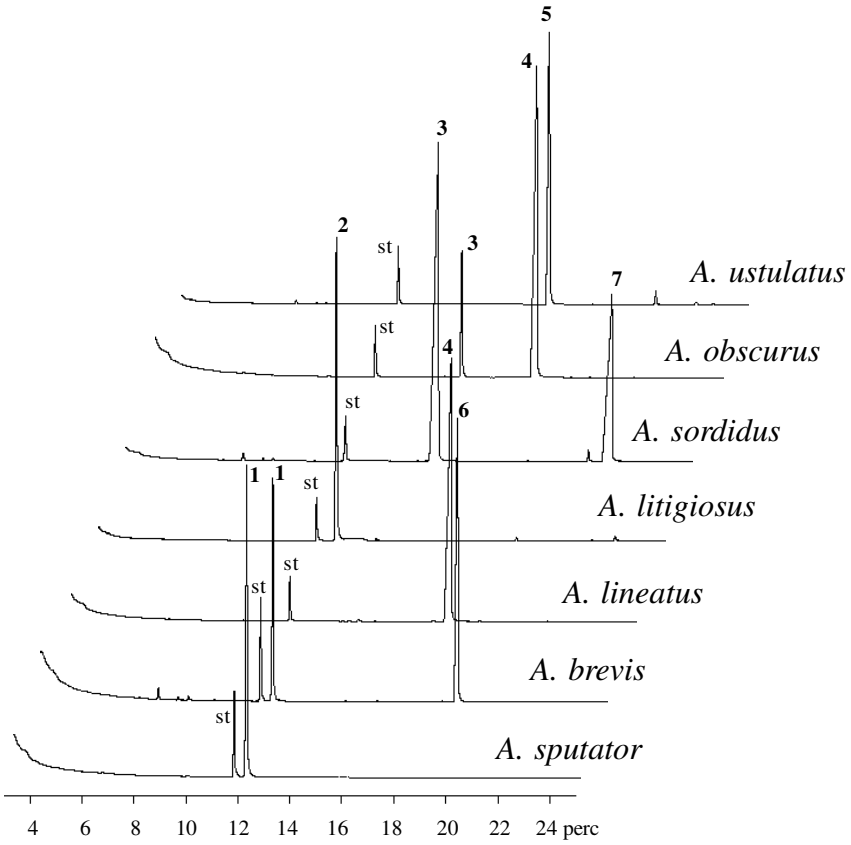
A. brevis

Concerning the pheromone composition of this species we did not find data in earlier literature. Our analyses showed that the pheromone extract was dominated by two components: geranyl butanoate and (*E,E*)-farnesyl butanoate (**1** and **6**, Figure 1). These same two compounds proved to be active in the field and the presence of both components was necessary for attraction of males to traps. A detailed description of the identification of the previously unknown *A. brevis* pheromone is given elsewhere (Tóth et al., 2002a). The optimized bait containing both components in equal amounts captured large numbers of A. brevis in Italy at all sites tested, with the exception of the Ligurian coast (Figure 2). Among other countries the presence of this species was reliably detected by our traps in Slovenia, Austria, and Bulgaria (near Sofia).

In Hungary, Romania and Croatia the bait was catching *A. sputator* probably due to the geranyl butanoate content (see also results and discussion for *A. sputator*). The same fact may explain catches of *A. proximus* Schwarz in Portugal (where no *A. brevi*s was caught), as at this site *A. proximus* catches were observed only in case of such baits which

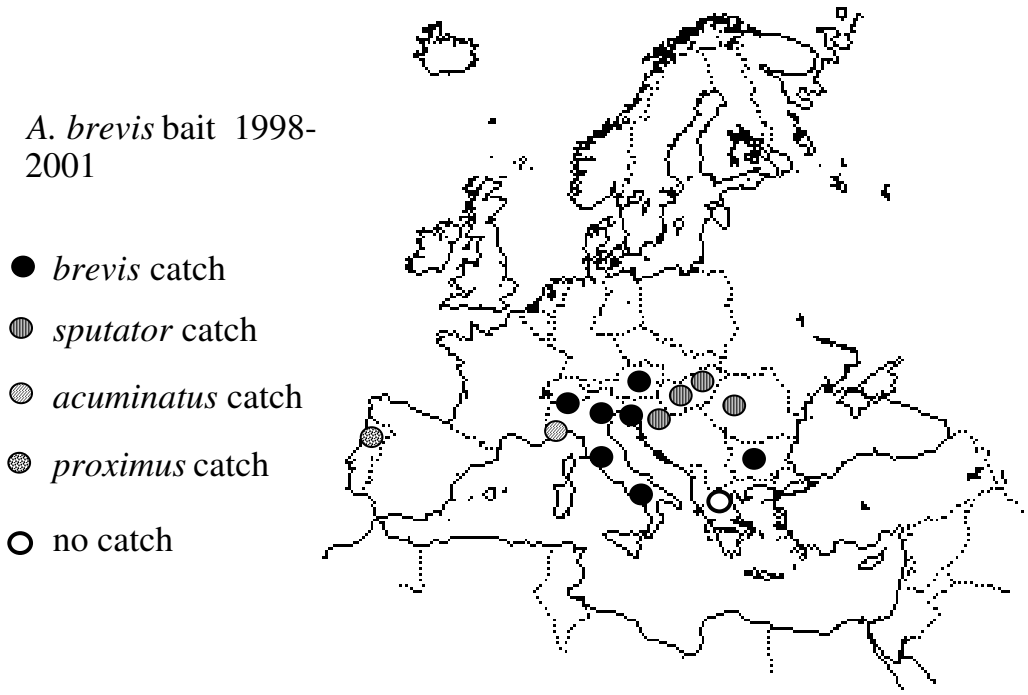
contained geranyl butanoate (alone or in combination; baits for *A. sputator* and *A. lineatus*).

Figure 1. Gas chromatographic analysis of pheromone gland extracts from female click beetles (*Agriotes* spp.)
Traces staggered by 1 min. Column; SP-2340 fused silica. **st** = internal standard (dodecyl acetate); **1** = geranyl butanoate; **2** = geranyl isovalerate; **3** = geranyl hexanoate; **4** = geranyl octanoate; **5** = (*E,E*)-farnesyl acetate; **6** = (*E,E*)-farnesyl butanoate; **7** = (*E,E*)-farnesyl hexanoate



This phenomenon needs further scrutiny, as the main pheromone component of Russian populations of *A. proximus* has been identified as (*E,E*)-farnesyl acetate, and several other farnesyl and geranyl esters (among them also geranyl butanoate) were present as minor or trace components (Yatsynin et al., 1996). In the field the 99:1 mixture of (*E,E*)-farnesyl acetate and neryl isovalerate was attractive (Yatsynin et al., 1980). Although this latter blend was not tested by us, it is noteworthy, that in our field tests in Portugal traps baited with (*E,E*)-farnesyl acetate did not catch a single *A. proximus*. Research for the discovery of the active pheromone composition for Western European populations of *A. proximus* is underway.

Figure 2. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. brevis* in different countries of Europe. Bait composition: geranyl butanoate / (*E,E*)-farnesyl butanoate in a ratio of 1:1



Catches of *A. acuminatus* Stephens at one of the Italian test sites may also be attributable to the geranil butanoate content of the bait. (see also discussion by *A. sputator*).

***Agriotes lineatus* L.**

The main component of the pheromone gland extract was found to be geranyl octanoate in our analyses (**4**, Figure 1). This compound has been previously described as the main pheromone component in *A. lineatus* by several authors (Borg-Karlson et al., 1988, Kudryavtsev et al., 1993, Siirde et al., 1993).

In our preliminary field activity test in Hungary in 1994 traps baited with geranyl octanoate caught a total of 30 *A. lineatus.* In the same test the mixture of (*E,E*)-farnesyl acetate and neryl isovalerate, described earlier as attracting *A. lineatus* populations in the West Ukraine (Kudryavtsev et al., 1993, Siirde et al., 1993) was inactive in Hungary. Instead of *A. lineatus*, this bait attracted 93 males of *A. ustulatus* (see detailed discussion later).

In a further preliminary test in Switzerland in 1997 the bait containing 10% geranyl butanoate added to geranyl octanoate (the main pheromone

component) caught a total of 273 beetles *versus* zero in traps baited with only the octanoate.
The presence of geranyl butanoate had been reported in this species, (Yatsynin et al., 1991, 1996), but no data on the activity of the binary mixture *versus* geranyl octanoate alone was published.
There was no significant difference between catches with the 10:3 and 10:1 octanoate:butanoate mixtures, and the 100:3 blend caught less at only one of the three sites tested.
Based on these results we used a 10:1 mixture in the Europe-wide comparative trials. Large numbers of *A. lineatus* were captured in almost all countries: United Kingdom, Germany, Austria, Switzerland, Italy, Slovenia, Croatia, Romania, Bulgaria, Greece and also at both sites in Hungary (Figure 3). No beetles were caught in Veneto, Italy, and in Portugal. In Portugal instead of *A. lineatus*, again catches of *A. proximus* were observed (see discussion by *A. brevis*).
Apart from Europe, our baits were successful in capturing *A. lineatus* also in Canada, where this species had been introduced probably from England (Vernon and Tóth, unpublished).

Figure 3. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. lineatus* in different countries of Europe. Bait composition: geranyl octanoate / geranyl butanoate in a ratio of 10:1

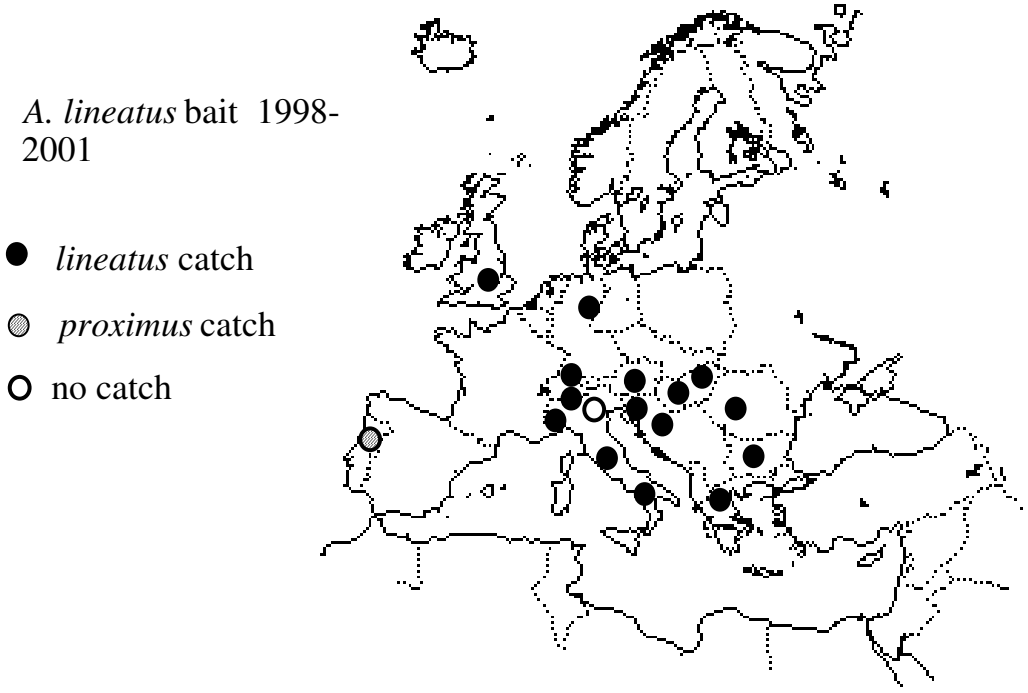
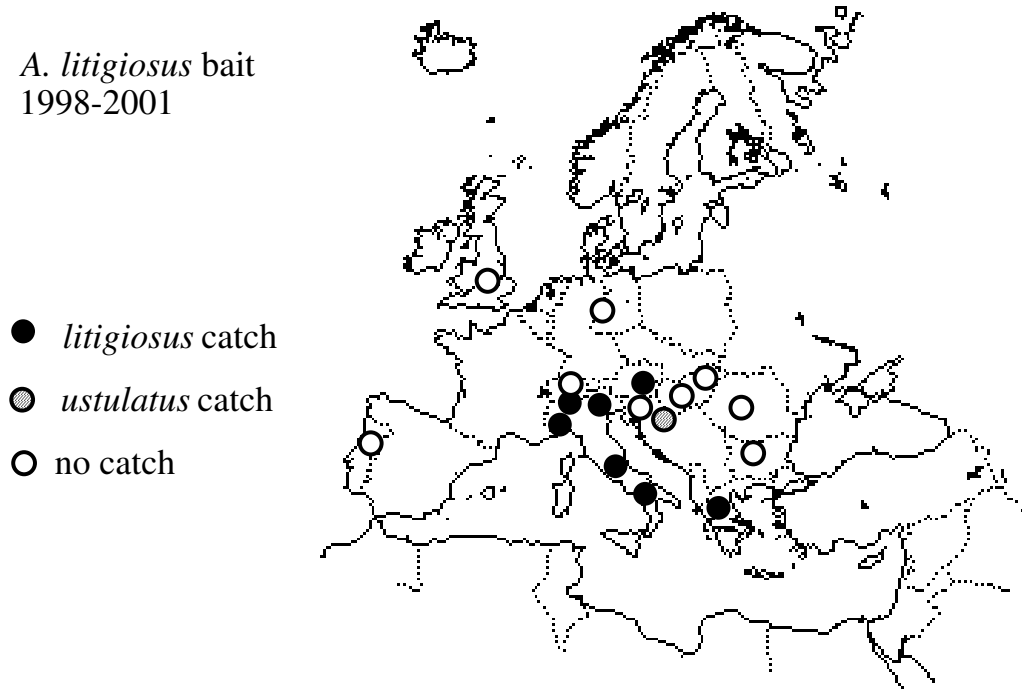


Figure 4. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. litigiosus* in different countries of Europe. Bait composition: geranyl isovalerate



***Agriotes litigiosus* Rossi.**

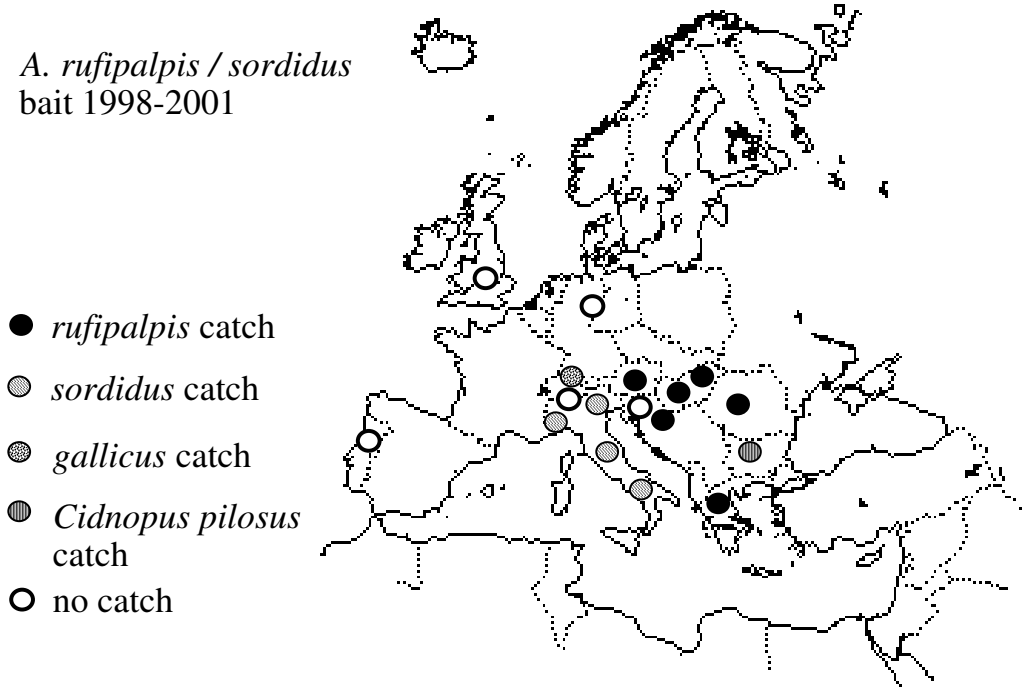
Russian authors reported geranyl isovalerate as the main pheromone of this species (Yatsynin et al., 1980, Kudryavtsev et al., 1993). However, later scrutiny revealed that these authors had worked with the species *A. litigiosus* var. *tauricus* Heyd. (V.G. Yatsynin, personal communication). Our results showed that the same compound occurred as the main pheromone component in *A. litigiosus* populations originating from Italy (**2**, Figure 1). There was no apparent difference between the pheromone composition of "dark" (= var. *laichartingi*) and "red" [= fenotypus (fen.) *typicus*] morphological forms of *A. litigiosus.* The two varieties, which are usually geographically separated, present differences in adult colour and larval morphology. According to observations conducted in Italy and Switzerland swarming patterns are different too (L. Furlan, personal communication).

The addition of (*E,E*)-farnesyl isovalerate or (*E*)-8-hydroxygeranyl 1,8-diisovalerate, two compounds which proved to be synergistic in *A. litigiosus* var. *tauricus* (Yatsynin and Rubanova, 1983) did not influence catches in any of the morphological forms of *A. litigiosus* (Table 4). Therefore traps

baited with geranyl isovalerate alone were used in the Europe-wide trapping tests.
The target species was caught in all Italian test sites, in Austria and Greece (Figure 4). At sites more to the north, so also in Hungary, no catches were recorded. On a single occasion some specimens of *A. ustulatus* were captured in traps in Croatia; however, since this result was not repeated, probably it was a result of cross contamination with pheromone samples during handing of the traps.

***Agriotes obscurus* L.**
Our analyses showed geranyl hexanoate and geranyl octanoate as dominant pheromone components in a ratio of 1:4 (**3** and **4**, Figure 1), supporting earlier reports on the presence of these two compounds in *A. obscurus* (Borg-Karlson et al., 1988, Yatsynin et al., 1996). Other terpene esters, such as the hexanoates and octanoates of nerol and 6,7-epoxygeraniol, could be detected only in trace amounts. Other terpenes detected were myrcene, limonene, *cis/trans*-ocimene, geranial and geraniol.
In contrast to earlier reports on the attractivity of geranyl hexanoate on its own (Kudryavtsev et al., 1993, Siirde et al., 1993), in our tests the presence of both compounds was necessary for attracting adults. No significant difference was observed between 2:1, 1:1 and 1:2 mixture ratios. Our present results support earlier findings in Russia (Yatsynin et al., 1996).
Traps baited with the above mixture captured large numbers of the target species especially in northern countries, or at sites with humid, cool climate, i.e. in the United Kingdom, Germany, Switzerland, Slovenia, Croatia and Romania (Figure 5). This bait was also very effective in Canada, where the species had been introduced probably from England (Vernon and Tóth, unpublished).
Instead of *A. obscurus* however, another species, *A. sordidus* Illiger was caught in italy, and *A. rufipalpis* Brullé in Greece (Figure 5). The geranyl hexanoate content of the bait may be an explanation of this phenomenon, as this compound is a potent sex attractant for both later spp. (see later).
It is worthwhile to note that in trappings in Hungary only sporadic *A. obscurus* catches were recorded in the course of several years of the present study, despite the fact that the bait proved to be highly effective at other places. This finding suggests that *A. obscurus* may be present in much lower population densities in Hungary than previously thought, and thus its pest status needs reinvestigation in this country.

Figure 5. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. obscurus* in different countries of Europe. Bait composition: geranyl octanoate / geranyl hexanoate in a ratio of 1:1



***A. rufipalpis* Brullé**

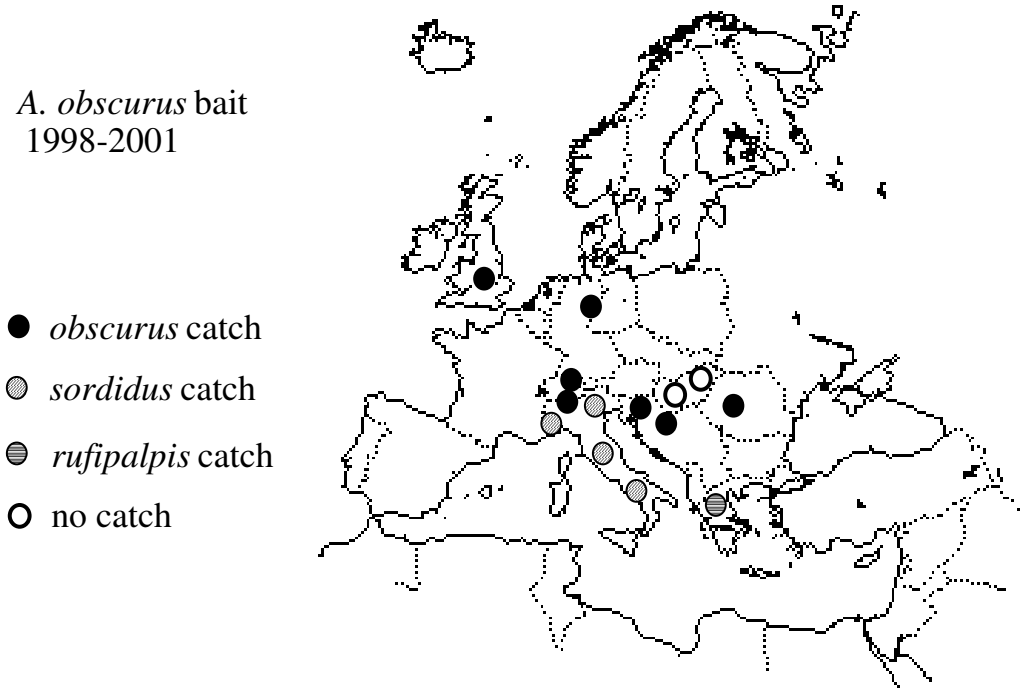
There was no published information on the pheromone composition of this species. In our studies no reliable analysis of pheromone gland extracts could be conducted, as we failed to collect female *A. rufipalpis* in large enough numbers. However, geranyl hexanoate was found to be attractive towards males of the species in the field (Tóth et al., 2002b). Traps baited with this compound captured well in Austria and Serbia. Especially high numbers were caught in Greece and Hungary (several sites), which suggests that the pest status of this species may be more important in these latter countries, than previously thought.

***A. sordidus* Illiger**

When testing the *A. rufipalpis* attractant geranyl hexanoate, large numbers of *A. sordidus* were captured in Italy (Tóth et al., 2002b). No previous information on the pheromone composition of this species was found in the literature. Analysis of gland extracts showed major peaks at the retention times of geranyl hexanoate and (*E,E*)-farnesyl hexanoate (**3** and **7**, Figure 1). Later field tests revealed that the presence of the farnesyl compound did not influence catches by the geranyl ester, which compound can be used

successfully alone for catching *A. sordidus*. Traps baited with this compound captured large numbers of males in all parts of Italy (Figure 6).

Figure 6. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. rufipalpis* in different countries of Europe. Bait composition: geranyl hexanoate



Based on our results it appears that *A. sordidus* is present only in the Western Mediterranean, while *A. rufipalpis* is widespread in the Eastern Mediterranean and Central Europe. In Slovenia, where the two areas may overlap, neither species was captured.

In Switzerland traps baited with gerany hexanoate captured *A. gallicus* Lacordaire, in Bulgaria, although in low numbers, *Cidnopus pilosus* Leske (Figure 6). The pheromone composition of neither species has been known before. Neither of them is regarded as a pest.

The fact that so many different species responded to geranil hexanoate in the different geographical regions may suggest that this compound – due probably to biosynthetic reasons – may possess a central position in the pheromonal microevolution of click beetles.

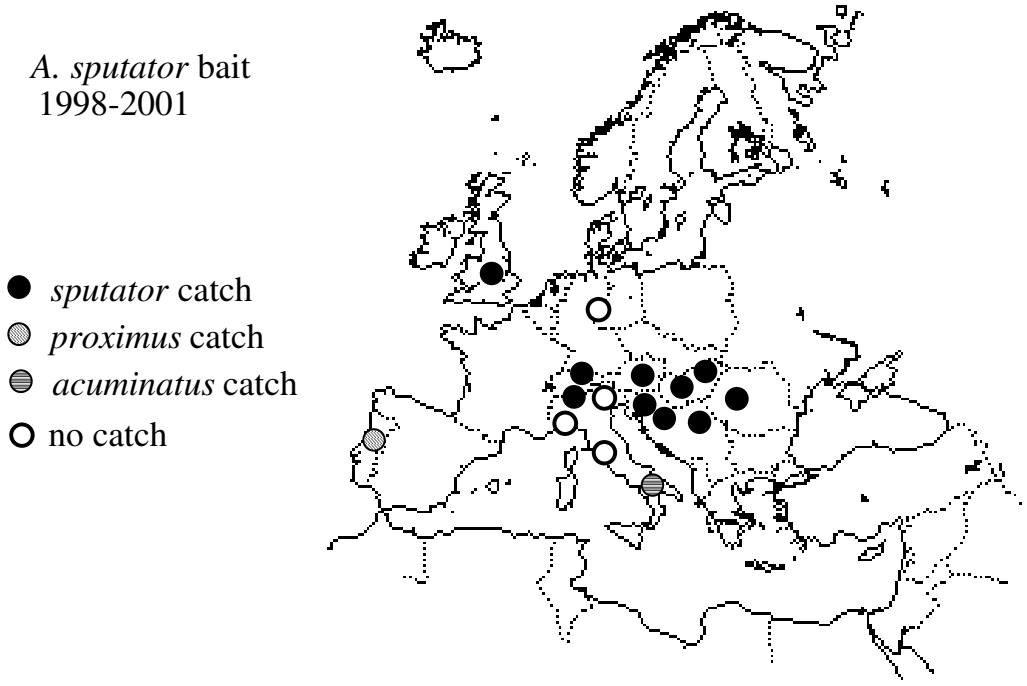
***A. sputator* L.**

In previous reports on the pheromone of *A. sputator* geranyl butanoate was reported as the main component (Siirde et al., 1993, Yatsynin et al., 1986). Indeed, results of our gland extract analyses showed that the extract was dominated by a very large peak of geranyl butanoate (**1**, Figure 1). Other compounds (some similar esters) were present only in traces.

In field activity tests in Hungary, geranyl butanoate on its own attracted large numbers of males and the addition of neryl butanoate, earlier claimed to be synergistic (Siirde et al., 1993), had no effect on captures. The addition of (*E,E*)-farnesyl hexanoate, alone or together with geranyl propionate also had no effect on catches by geranyl butanoate, although these compounds had been reported earlier to be present in pheromone extracts (Yatsynin et al., 1996). Consequently we used geranyl butanoate on its own as a bait for the Europe-wide trapping tests.

Most beetles were caught in countries to the north and in Central Europe – United Kingdom, Switzerland, Serbia, Romania, Bulgaria, Austria, Slovenia and Hungary (Figure 7). Although we did not observe catches in the trial in Germany (near Berlin), probably only local lack of a high enough population may be the cause for this phenomenon.

Figure 7. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. sputator* in different countries of Europe. Bait composition: geranyl butanoate

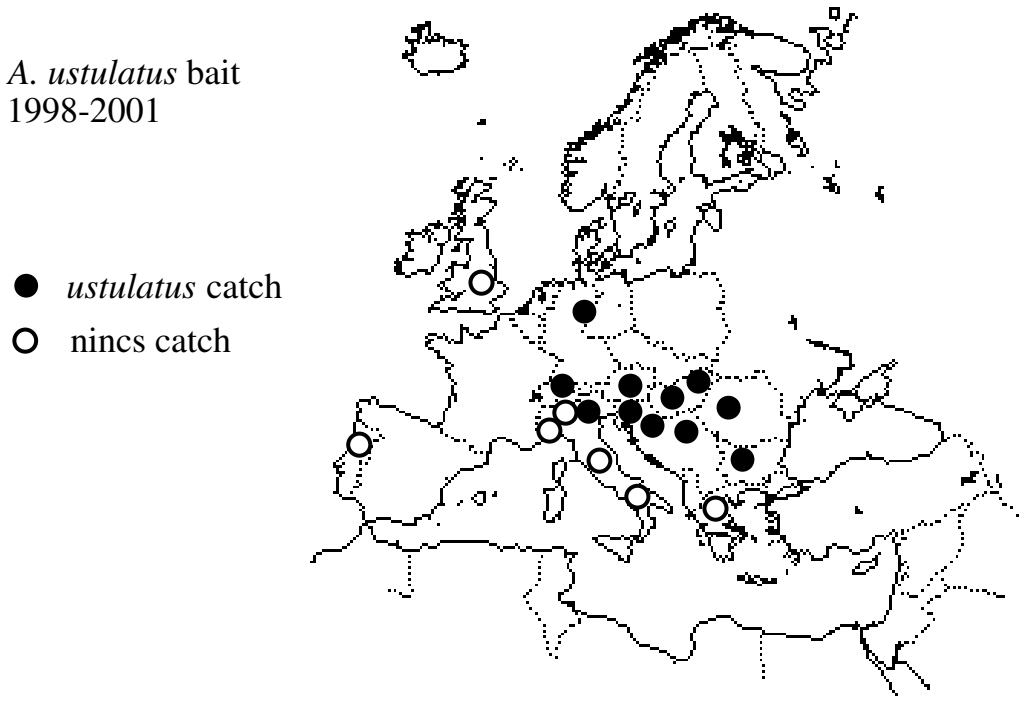


Our bait was excellent in capturing *A. sputator* also in Canada, where the species had been introduced from England (Vernon and Tóth, unpublished).
At one of the Italian sites we recorded catches of *A. acuminatus*. Probably geranyl butanoate is a sex attractant for this species, as it was also captured in traps with the *A. brevis* bait (which also contains this compound), also in Italy. There is no mention of the pheromone composition of this species in the literature. The species is not regarded as an important agricultural pest.
In the tests in Portugal, where no *A. sputator* was caught, the traps regularly captured *A. proximus*. This result inevitably points to the importance of geranyl butanoate in the pheromonal communication of this species (see also by *A. lineatus*).

***A. ustulatus* Schaller**

In pheromone extracts of *A. ustulatus* females originating from Italy our analyses showed (*E,E*)-farnesyl acetate to be the predominating component in the pheromone gland extract (**5**, Figure 1), confirming earlier results for populations in Russia (Kudryavtsev et al., 1993, Siirde et al., 1993, Yatsynin et al., 1996).
There were virtually no differences in pheromone composition of extracts from the "black", or "red" morphological phenotypes, which are always present in any population, irrespective of source. Consequently we used (*E,E*)-farnesyl acetate on its own in the bait for our Europe-wide trapping efforts.
High captures were recorded in Germany, Switzerland, the northeastern part of Italy, Austria, Slovenia, Croatia, Serbia, Romania, Bulgaria and Hungary (Figure 8). No captures were recorded in the United Kingdom, Portugal, Greece, and other parts of Italy.

Figure 8. Click beetle spp. captured in traps baited with the synthetic pheromone of *A. ustulatus* in different countries of Europe. Bait composition: (*E,E*)-farnesyl acetate



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Summary

EUROPE-WIDE PHEROMONE STUDIES ON CLICK BEETLES (COLEOPTERA: ELATERIDAE)

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In the course of the above, Europe-wide trapping tests using the pheromone baits discovered or optimized by our team we were successful in showing out the geographical occurrence of the 8 most important agricultural pest click beetle species. The application of pheromone traps shows perspectives in the detection, monitoring and establishment of damage thresholds for these species, or in some cases in the direct control through mass trapping.

Acknowledgements The authors are indebted to all cooperators who participated in the above studies at the different localities. This research was partially supported by grants OTKA T017693 and T029126, and also by grant NKFP OM-00116/2001.

EXPERIENCE OF THE DEVELOPMENT OF NEW BROAD SPECTRUM MICROBICIDE FOR PLANT PROTECTION. THE ROLE OF SYNERGY

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Enhancing pesticide action through mixtures is a powerful tool for improvement of current pest control technologies. Numerous models have been constructed for evaluation of the character of joint action of chemicals (Bánki, 1978), which mixtures meets special problems of assessment and data treatment. Joint action of compounds *A* and *B* can be performed by different manner:

- Additive or aggregative effect; the total effect is a simple sum of the proper effects of parties, i.e., A+B=C, where the expected effect C is equipotent to the measured effect (ME) of their mixture. In this case $|C-ME| < LSD_{0.05}$ of analysis, and one can assume that the biological effects of the parties do not interfere.
- Antagonism (counteraction) opposition in biological action; interaction of two or more substances such that the action of any one of them on organisms is lessened, i.e., A+B=C, where C > ME, and $|C-ME| > LSD_{0.05}$.
- Synergism; interaction of compounds such that the total effect is greater than the sum of the individual effects, i.e., A+B=C, where C < ME, and $|C-ME| > LSD_{0.05}$.

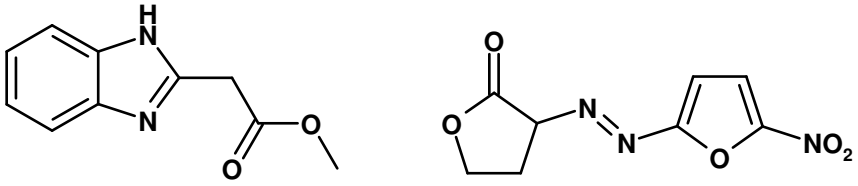
Two basic standpoints should be considered in decision process of product development when patents support monopoly rights:

The novelty of a finding is an absolute criterion, however there is relatively easy to find nowadays an earlier publication canceling the patent application. While the obviousness is a complex problem, and it relates a number of headings, inventive level, inventive step and technical advance. The first two aspects meet very subjective criteria. What seem to be masterfully inventive to one person can appear evidently obvious to another. However the technical advance can be supported by experience as well as by comparative analysis to present level of technology.

The optimized mixtures of nitrofurane and benzimidazole derivatives efficiently inhibit large number of microbial species (Ángyán *et al*., 1990; Detre *et al.*, 2000) particularly streptomycin tolerant bacteria and benomyl tolerant fungi (Oros et al., 2001). Basing on the example of these molecules

we demonstrate the steps of decision process (details of toxicological methods are published in cited papers):
(i) Qualitative analysis of interaction using agar diffusion technique or comparative analysis dose/response lines for facultative and obligate parasites, respectively, (ii) optimization of the mixture applying the model of Horsfall and Dimmond for determination of the ideal mixing ratio as well as the Sun's model for calculation of the rate of synergy, (iii) characterization of the spectrum of activity of new preparation.
Furazolidone (Figure 1), formerly used in veterinary, significantly and selectively inhibits (MIC < 25 mg/L) phytopathogenic *Erwinia* and *Xantomonas* species (Ángyán *et al.*, 1990), while carbendazim (Figure 1) is well known agricultural fungicide of systemic activity. The antimicrobial activity of their mixture surpass
benomyl and streptomycin against fungi (*Cochliobolus*, *Cytospora*, *Valsa* and *Ustilago*) as well as bacteria (*Erwinia* and *Xanthomonas*). The optimized mixtures efficiently inhibit streptomycin tolerant bacteria.

Figure 1. Structure of synergically interacting compounds.

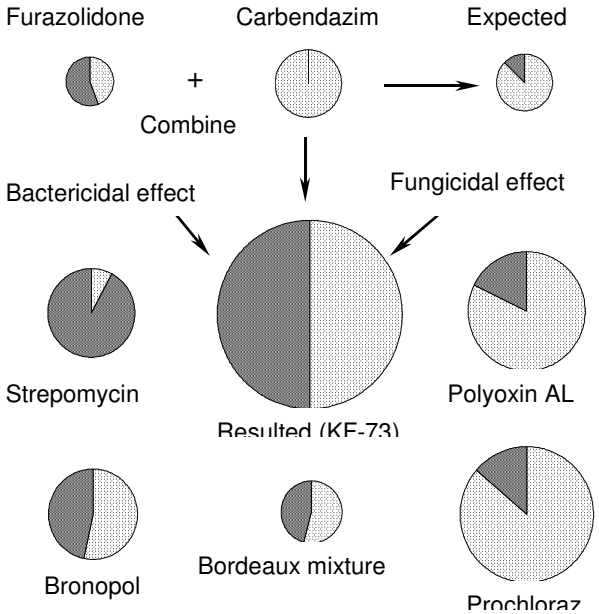


Carbendazim Furazolidone

The synergic joint action between these substances was first detected by agar diffusion technique testing activity against a strain of *F. oxysporum* tolerant to benomyl (Hornok *et al.*, 1988), and was later verified against pathogenic *Rhizobium radiobacter* (str. C58) as well. The increasing economic importance of phytobacterioses compels to discover new methods for disease control. The worldwide incidence of acquired tolerance to agromicrobicides pressurizes also on research and development. Research is usually paid for out of the benefits made from the utilization products of research. As the data mentioned above were inspiring and the mixture was optimized and it's spectrum of activity was assayed (Figure 2). Results indicated significant increase of microbicidal potency of the mixture that impressed further speculations on development. The superiority of opened model of development for this case was also shown by preliminary cost/benefit calculation (Figure 3) supporting efforts for the enhancement of

pesticide action through mixtures for improvement of current pest control technologies.

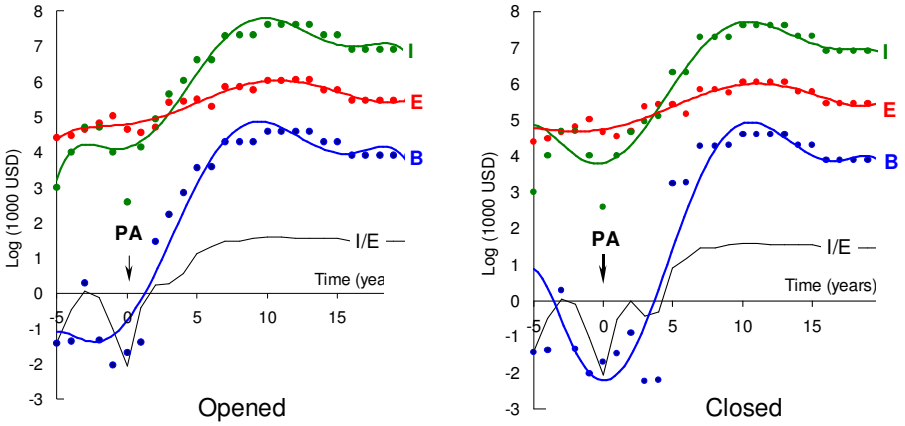
Figure 2. Joint action of furazolidone and carbendazim against pro- and eukaryotic microbes.
The area of circles is proportional to overall antimicrobial potency of compounds, while dotted and closed areas represent bactericidal and fungicidal potencies, respectively.
Potential activities were calculated by Potency Mapping (Lewy, 1976) based on activity of compounds against 25 bacterial and 42 fungal species, respectively.



As it is shown both increase of incomes and return of investment are more rapid by following the opened developmental strategy.
According to requirements for patentability the combined action of parties must be greater than the sum of action of each of them used alone at the same mass. It means that as minimum as 210 experiments should be carried out when applying the graphic model for establishment of most potent variant. The ideal weight ratio of carbendazim and furazolidone was determined according to Horsfall and Dimmond (1941).
This model makes possible to decrease the size of laboratory work requested to generation of acceptable and convincing data for patent application.

Figure 3. Financial effectiveness of various strategies in pesticide development.

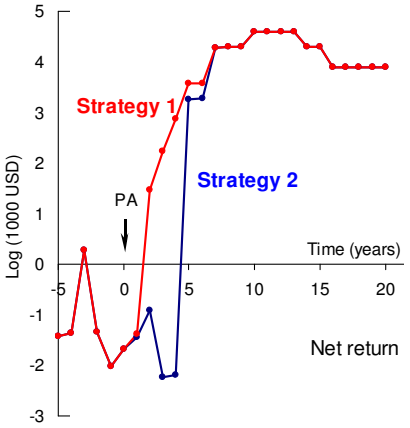
PA= patent application, I= incomes, E= expenses and B= benefit. The calculation was made on the example of development of new agricultural fungicide KF-73, where 40 USD/kg of commercial price of preparation was considered.



Strategy 1.

Results of the work are public and inventors are independent on manufacturer and investors.

Researchers are competing and program managers serve to inventors.



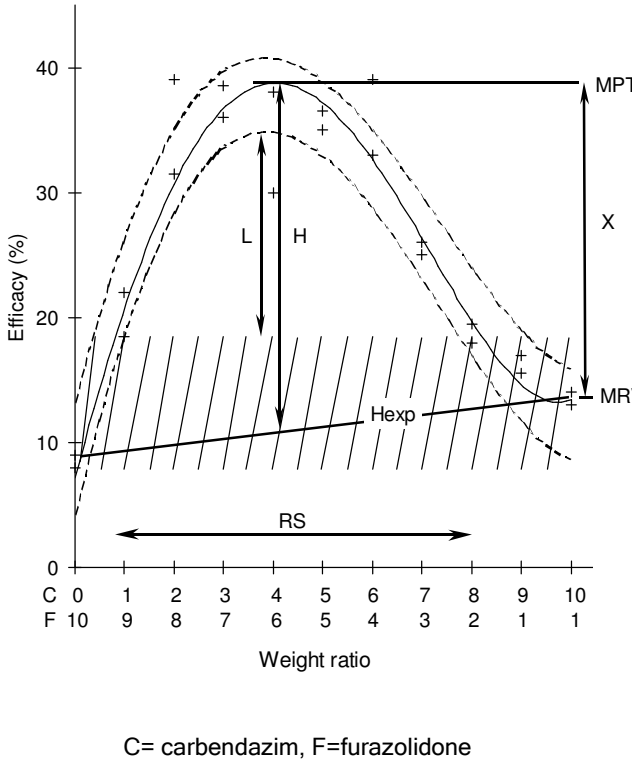
Strategy 2.

Results of the work are closed of public and inventors depend on manufacturer and investors.

Investors are competing and inventors serve to program managers.

Moreover there is possible to decide more rapidly on the followings due to shortening the period of data collection as 80 experiments supply the suitable results. We have made some modifications of this model, which allow to evaluate the statistical significance of the range of synergy (Figure 4).

Figure 4. The optimization of weight ratio of mixed compounds
Test organism: *Bipolaris zeicola* (Stouth) Shoem.
Dose: 10 mg/L
Medium: PDA

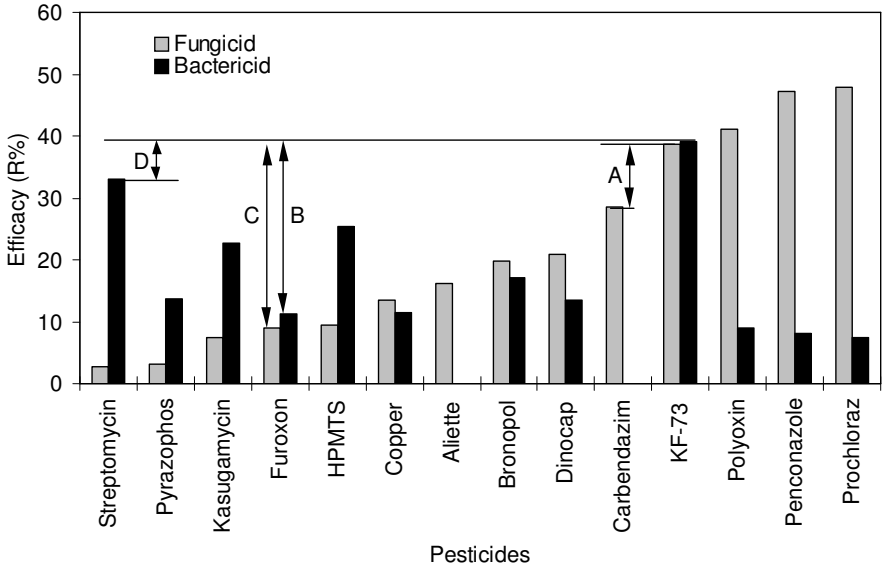


Parameter:
inhibition of the linear growth of hyphae
RS – range of synergy
Hexp – expected activity according to Horsfall's model
MPT – Most potent treatment
MRV – Maximum response value
Synergic increase of activity:
X=MPT-MRV – demanded by Worley *et al*. (1969)
H=MPT-Hexp
$L=LC_{MPT}-UC_{MRV}$ at $P_{0.05}$ is the verified significant shift.

The new model is more strait and determines the range of synergy more narrowly as the sections of activities of lower confidence limit (LC) for the mixture and upper confidence limit (UC) for the more potent agent are used as boundary values. Besides of the more strict approach to the mixing range, the rate of synergy is also evaluated more accurately determining it as the difference
$L=LC_{MPT}-UC_{MRV} > LSD_{0.05}$. The Horsfall's model originally measured the difference between the response to most potent treatment (MPT) and that expected by the model (Hexp) and this value (H) is higher than the rate requested according to demands by *re Lemin*'s case (Worley et al., 1969) where the efficacy of the mixture should surpass the activity of more potent partner (MRV), i.e., MPT > MRV, obligatorily (see Figure 3). According to our proposal L<X<H as MPT>MRV>Hexp. Although we have less

variability of the composition but as a result the strength of decision has increased.
It was verified that the spectrum of antimicrobial activity of mixture is broader and it is more active than either carbendazim or furazolidone (Figure 5). The overall bactericidal activity of the optimized preparation (KF-73) surpassed the streptomycin and kasugamicin, and it was far more potent than the synthetic agrobactericides.

Figure 5. Comparative analysis of the antimicrocial spectrum of activity



Columns are proportional to potency values calculated from response data of 42 fungal and 25 bacterial species, respectively.

The antifungal activity of new preparation (Pharmaplant 40 fw) is better than that of benomyl, the most potent member of benzimidazole series of fungicides. Moreover, it is highly active against strains tolerant to benzimidazoles (*Botrytis*, *Fusarium*, *and Venturia*). Although some azole derivatives (prochloraz, penconazole) exhibit higher antifungal potency but their therapeutic value is lower.
The therapeutic index (T.I), - the ratio of Maximium Tolerated Dose by host plant and Minimum Inhibitory Dose for parasite, - is an important measure for determining the area of use of any agromicrobicide. Unfortunately, azole derivatives cause severe stunting even in their therapeutic doses, which limits the area of their use. Pharmaplant 40 fw free of this disadvantage. Its bactericidal activity is probably equivalent to streptomycin (*Erwina*,

Xanthomonas), however it acts against streptomycin tolerant strains as well. Moreover, *Pseudomonas fluorescens* and *Pantea agglomerans*, the important natural antagonist of phyotpathogenic fungi, tolerate well the new microbicide.

The probability of the appearance of bacterial strains with acquired resistance to Pharmaplant 40 fw was lower than 10^{-9} as determined on *E. carotovora*. Neither carbendazim nor furazolidone proved to be mutagenic in agrocin/*Rhizobium radiobacter* test (Oros 1983).

It was verified that the spectrum of antimicrobial activity of optimized mixture is broader and it is more active than carbendazim and furazolidone, in particular against species affiliated to *Erwinia* and *Fusarium* genera. The significant enhancement in activity of mixtures with other microbicides makes possible weighty decrease in applied doses leading to reduction the time of tolerance and costs of pest control. The applied models of data treatment are suitable for estimation of changes in efficacy of mixtures, and the results stand all demands required by US Patent Office for synergy.

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SAVE ALTERNATIVES OF METHYL-BROMYDE FOR CONTROLLING SOIL-BORNE PATHOGENS INFESTING EXPORT VEGETABLES IN NEWLY CLAIMED DESERT LANDS IN EGYPT

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(Manuscript has not arrived)

ACTUAL PROBLEMS OF PLANT PROTECTION IN HUNGARY

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The major challenge in the plant protection practice is the corroboration the basic methods of integrated pest management; the introduction of novel, environmentally safe and sound methods; the development of sustainability in the agricultural practice. By the distribution of different commercial goods of agriculture new pathogens spread, new pests were introduced and a lot of new weeds became abundant in the country. All of these harmful effects treat the safety and the quality of agricultural production.

In the frame of this paper the authors would like to summarize the relatively new or economically most important pests, pathogens and weeds, against which the common plant protection measures were still not successful or remained to be unsolved.

Insects

In 2003 the most important insect pests, causing serious yield losses were the following: corn weevil (*Diabrotica virgifera virgifer*a), corn earworm (*Helicoverpa armigera*), western flower trips (*Frankliniella occidentalis*), rootworm nematodes, and flea beetles (*Phyllotreta* spp.).

The American corn weevil occurred in some new counties, especially in Zala County, in the Transdanubian area (Ripka et al. 2000). The most serious infestation occurred in counties Tolna and Baranya. New control methods were worked out, among them it seemed to be the more successful the soil insect technology, and the chemical control of adults 8-12 days after the migration peak.

Helicoverpa armigera occurred in great mean numbers. It was observed in almost all field crops and vegetables in all parts of the country. Damages were detected in sunflower and sweet corn plantations as well as in tomato and green peppers (Szeöke and Dulinafka 1987, Vörös et al. 1997). Overwintering conditions of *Helicoverpa armigera* are still not known, but studies on this field has started.

The western flower trips cause damages in every year, both in the land and in the greenhouses. Serious problems were estimated in green pepper and tomato production, as well as in ornamental plants especially in roses and carnations. In addition to the conventional methods, new biological control methods were increased with the use of predatory mites (*Amblyseius* spp.) and predatory bugs (*Orius* spp.) (Budai 1986, 2002).

Every year, the damages caused by rootworm nematodes (*Melodygine* spp.) mean great problem in vegetable production, both in the field and in the greenhouses. The problem will increase when the use of methyl bromide is prohibited. The hot and dry weather of 2003 increased populations of pests and damages caused by flea beetles in cabbage and carrot plantations and *Phyllotreta vittula* in maize.

Pathogens

Among the viruses *Potato virus Y* (PVY) remained the most important field pathogen, causing significant yield losses both in potato and tobacco production. The virus has many strains (PVY^{O}, PVY^{C}, PVY^{An}) which were described in Hungary by Horváth (1966, 1967a,b). Among them the venial necrotic strain (PVY^{N}) caused significant damages in tobacco fields in the latese years, in which moisture and temperature were favourable for the large-scale distribution of winged aphid vector species. The tuber necrotic ringspot strain of PVY (PVY^{NTN}) was described first in Hungary, and nowadays distributed in all over the world. This resistance breaking strain overcame the resistance of the commercial varieties and became the most abundant PVY strain in Hungary (Takács et al., 1998, 2003). There are only three possibilities for the control: first useage of resistant varieties; direct chemical control against aphids; monitoring of winged vector species in the fields and taking agricultural control measures, like isolated production of *Solanaceous* crops (pepper, tomato, potato, tobacco). It is a fortune that there are available six Hungarian tobacco varieties, which are resistant (immune) to PVY strains.

The other, economically important pathogen is the *Tomato spotted wilt tospovirus* (TSWV). This virus vectored mainly by two trips species: the endemic onion (tobacco) trips (*Thrips tabaci*) and the adventive western flower trips (*Frankliniella occidentalis*) (Jenser et al., 1996, Jenser and Gáborjányi 1998). The pathogen has been known as tobacco pathogen since 30 years. In the past years by the distribution of *F. occidentalis* TSVW spread in tomato and pepper cultures, grown under plastic tunnels and greenhouses (Gáborjányi et al., 1994). The problem of spotted wilt disease

was successfully reduced by the new, uniform technologies of seedling production (floating bed method), and by the direct monitoring of the two vector species both in the greenhouses as well as in the fields (Gáborjányi et al., 2002, Fekete et al., 2003, Jenser et al., 2003).

Stolbur disease, caused by the *Stolbur phytoplasma* (SP) caused only local, sporadic epidemic, however the economic loss was valuable in some years, when the dry and hot wheather was suitable for the distribution of vector species (Gáborjányi et al., 1998). Tomato, tobacco, pepper, and more recently grapes were suffering of the SP infection, but in this year (2003) a massive infection of potatoes were also observed. Monitoring would easily control the abundance of leafhopper species, but as we know no efforts have been made on this field.

The bacterial blight of pome fruits (*Erwinia amylovora*) remained a permanent, and still unmanaged problem. "Fire blight" occurred in every year, when the temperature and the moisture were enough high for the infection in the blossom period of pear and apple trees. *E. amylovora* is still a quarantine pathogen, but we have to understand, that it was established in our ecological conditions and the control measurements were not sufficient to overcome the infections. The drastic cuts of the infected parts and the use of different antibacterial chemicals including antibiotics, still not effective to reduce the infection.

Brown rot disease, or Southern bacterial wilt of potato caused by *Ralstonia (Pseudomonas) solanacearum* is a recently emerged, extremely destructive disease in Hungary. The bacteria had been dragged by seed potato import from the Netherlands. The useage of rapid and effective quarantine measurements were successful in the suppress of this harmful, ecologically important pathogen. The pathogen is still not generalized in Hungary but based on the former bad experiences we can calculate possible occurrence of new infections too.

Grapevine plantations are seriously affected by the early decline of grapes. The early death of grape stocks caused by different pathogens, including viruses (*Grape feanleaf virus, Grapevine stem pitting virus*), bacteria (*Rhizobium /Agrobacterium/ tumefaciens*), and fungi as *Eutypa lata* or *Phomopsis viticola*. The aetiologically complex disease, commonly named ESCA could be cause serious yield losses in the plantations. The potentially involved pathogens are the followings: *Stereum hirsutum, Phellinus (Fomitiporaria) igniarius, Phaeommoniella chlamidospora* and *Diplodia mutila*. The control is very difficult and hopeless, because of complexity of causal agents.

2003 can be looked conflicting in crop production. This year weed control could be extraordinary regarded. Unusual dryness and lack of precipitation could be sensed in the late spring. There was not enough precipitation to the efficacy of the pre-sowing and pre-emergence herbicide treatments, especially in the hoed crops. The importance of the early post-emergence treatments, which were not negligible previously, either were increasing, especially on strongly weedy areas. Among the weed problems we would like to deal only with those species that cause bigger and bigger damages.

Nowadays *Ambrosia artemisiifolia* L. is the most widely spreaded weed species in Hungary. Common ragweed produces a lot of pollens with allergenic effect so it causes mass human diseases. It had been dragged from North America and belonged to those weeds, which very quickly spreaded and had mass appearance in the past decades. Common ragweed planted itself to Hungary in the early twenties of the 20th century. The infected area was more than 380.000 ha in 1996 (Béres et al. 1998). Based on the data of the 4th National Weed Survey, the common ragweed is one of those weeds that can be found the most frequently in Hungary. In 1950 it was on the 21st place, in 1970 it had the 8th place, and in 1988 it was on the 4th place among the weed dominance both in winter wheat and maize cultures, and arised for the 1st place by 1997 (Béres, 2003).
The spread of common ragweed is not a new problem. It has several biological characteristics, which help its spreading and survival. It is known that the seeds preserve their germinate ability for 30-40 years in the deeper layers (35-45 cm) of the soil. Common ragweed has tolerant and resistant biotypes against triazine herbicides (Béres et al., 1998).

Datura stramonium L. is an annual weed species. It does not spread vegetatively; it can overwinter with seeds in different layers of the soils only. 50 years ago Ujvárosi (1973) ranked it on the 109th place of the weed dominance order of the 1st National Weed Survey in maize. Twenty years later (1969-71) jimson weed was on the 36th place. Based on the 3rd National Weed Survey it was occurred on 585 737 ha and the heavily infected area was 231 612 ha. 6 601 ha were highly infected, 18 192 ha were medium infected area, and 59 076 ha were mild infected (Tóth et al., 1998). On the 37.95% of infected areas produced maize, 18.22% sunflower, and 15.74% other plants, respectively (Czimber and Hartmann, 1998).
Jimson weed is spreading not only in Hungary but also in other countries of the world. It appears especially in soybean, bean and maize. The tolerance

against carbamide herbicides has an important role in its spreading. The damage of Jimson weed can be similar to the quantity of its spreading (Czimber and Hartmann 1998).

Hemp (*Cannabis sativa* L.) as a weed causes serious problems in Hungary on arable lands and uncultivated areas. Results of National Weed Surveys show that hemp covers increasing areas both in cereals and hoed crops. In the past years the increase and spreading of hemp is conspicuous on bigger and bigger areas (Benécsné, 2002, 2003). Its damage can be significant because it dwarf the sowing and deprive the crop of water and nutrients (Lehoczky and Reisinger). Because of its great fresh weight and massive stem it makes heavy the harvest.

From the point of view of weed control perennial weeds mean especially great problem that can be solved with integrated protection only. Based on the 4th National Weed Survey (1996-1997) data it takes the fifth place in the dominance sequence, with 1.8% average covering (Tóth and Spilák, 1998). Intensive nutrient uptake has an important roll in considerable competitive capacity of *Cirsium arvense* especially in competition for nutrients (Lehoczky et al., 2003).
In spreading this weed plays an important role its capacity of vegetative reproduction. General use of herbicides a little bit repressed the Canada thistle but nowadays the importance of this weed is increasing again. Canada thistle is wide-spread in all territory of Hungary; it can be found almost on every soil types. This plant is one of the most difficulty-eradicated weeds of lands, ruderals, and roadsides (Benécsné and Hartmann, 2002).

The average cover of reed (*Phragmites australis* /Cav./ Trin. ex Steudel) has considerably increased compared to the earlier years. In 1952 common reed was on the 32nd place of the weed dominance order with 0.1857% weed cover. It had the 42nd place with 0.1055% in 1964 (Németh et al. 1998). Common reed has got into the 12 mapped weeds and this indicates the spreading and becoming an important weed species. In 1987 the infected area was 163 000 ha, and 188 000 ha in 1989. The 40% of the infected areas were cereals and 50% were maize and other hoed crops. From the 2nd National Weed Survey (1969-71) common reed appears in increasing quantities in maize. The infection of Hungary have two centres. It is coming from the direction of West or East and its average cover is increasing. Its spreading is heavy in Vas, Somogy, Fejér and Tolna counties. In Heves County its cover did not reach 0.083 %, and at the last weed survey its cover exceeded 0.56 % (Lukács, 2002).

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LECTURES OF PHYTOPATHOLOGICAL SESSION

SURVEY OF VIRUS DISEASES OF TOBACCO FIELDS ON THE EASTERN HUNGARY

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Eastern Hungary, counties Szabolcs-Szatmár-Bereg and Hajdú-Bihar were always traditional territories of tobacco production. Both yield safety and yield qualities were significantly dependent on the severity of virus infections. Formerly *Tobacco mosaic virus* (TMV) and *Cucumber mosaic virus* (CMV) were abundant, later the tuber necrotic ringspot strain of *Potato virus Y* (PVY^{NTN}) caused serious yield losses. Recently PVY, and *Tomato spotted wilt virus* (TSWV) became to the most important viral pathogens in this region (Gáborjányi et al. 2001, 2002; Horváth et al. 2001). Epidemic of these two virus diseases is always consequence of the rapid reproduction and migration of viruliferous aphid and trips species. Neighbourhood of other solanaceous plants, especially the large-scale production of potato also increase the infection pressure. The use of modern environmentally safe and sound methods of integrated plant protection needed the permanent monitoring of virus occurrence. The aim of the present work was to follow the quality and abundance of virus infections in tobacco fields.

Materials and Methods

Survey of virus infestation of tobacco fields were made according to the visual symptoms. Symptomatic leaf samples of tobacco and potato plants were collected from the fields in the district of counties Szabolcs-Szatmár-Bereg and Hajdú-Bihar. Frozen samples were homogenized in a five volumes of buffer solution according to the recipes of Clark and Adams (1977). Virus content was measured by DAS ELISA test using antisera of Loewe Biochemica Ltd. in a Labsystem Multiscan RC ELISA reader at 405 nm wavelengths. Samples were considered positive, if their absorbance

values exceeded three times than those of healthy control ones. In the liability tests infections of *Alfalfa mosaic virus*, (AMV), CMV, PVY, TMV, *Tomato mosaic virus* (ToMV), and TSWV were studied.

Results and Discussion

Visual survey of virus infestation of tobacco fields in 2003

Occurrence and distribution of viruses in the tobacco fields were estimated by visual observations according to the symptoms developed on diseased plants (Table 1).

Table 1. Incidense of the most important tobacco pathogen viruses on the eastern Hungary

Variety	Viruses			
	PVY	TSWV	CMV	TMV
Hevesi 9	0.0	0.1	0.1	0.0
Hevesi 17	0.0	0.0	0.3	0.1
Hevesi 18	0.0	0.0	1.0	0.0
Pallagi 1	0.3	0.4	0.1	0.1
Pallagi 3	0.6	0.4	0.0	0.0
Pallagi 5	0.0	0.3	0.2	0.0
TN-86	0.0	0.0	0.0	0.0
TN-90	0.0	0.3	0.0	0.0

The abundance of virus infections in 2003 remained low, in comparison to the former years. Massive gradation of the virus vectors, both of aphids and trips, did not develop because of the very dry and hot wheather in spring and summer. PVY infection, based on the veinal necrotic symptoms, occurred only in two Hungarian Burley varieties (P-1 and P3), but no infection was detected in flavouring Burley varieties (TN86 and TN90). Similary to PVY the infection ratio of TSWV was relatively low, ranged from 0.1- 0.4 %. Some Burley varieties (P1, P3 and P5) were relatively susceptible to TSWV infection. CMV, formerly the most common virus in tobacco fields, remained in a low level, only one Hungarian variety (H-18) showed relatively higher infection rate. No or very low infection of TMV was detected by visual observation because of the general resistance of the used varieties to TMV. In contrast to the former years, in 2003 no complex infection was estimated. The reduced level of natural infection gave no possibilities to compare the susceptibility or resistance of the different varieties.

Testing the liability of visual observations by ELISA method

Liability of the visual observations was checked by serological methods. Diseased plants bearing typical symptoms were collected and the virus content were measured by DAS ELISA serological tests. Comparison of the two diagnostical methods is in the Table 2. Results showed that the TSWV has the most characteristic symptoms, and all isolates correctly judged to TSWV. Only in two cases samples were free from TSWV. Typical mosaic symptoms of PVY, even veinal necrotic ones, could be mischaracterized. In one case a complex infection of PVY and TSWV occurred. Similarly the so-called mosaic symptoms could be due not only by TMV, but also CMV or PVY infection. Mottling of the leaves, and oak leaf pattern were well characterized as typical symptoms of CMV. These comparison shows, that visual characterization of virus diseases is very important, but not sufficient for testing plant viruses.

Table 2: Testing the liability of the visual observations by serological tests

Pleace	Variety	Symptoms	Virus suspected by visual symptoms	Virus found by ELISA
Bököny	Pallagi 3	M	TSWV	TSWV
Bököny	Pallagi 3	M, Mo	TSWV	TSWV
Bököny	Pallagi 3	M	TSWV	-
Bököny	Pallagi 3	M, Ldef	TSWV	-
Bököny	Pallagi 3	Vn, Chl	PVY	TSWV
Bököny	Pallagi 3	Rs	TSWV	TSWV
Bököny	Pallagi 1	M, Vn	PVY	PVY, TSWV
Debrecen	Virginia type	Vc	PVY	TSWV
Debrecen	Virginia type	Vn	PVY	PVY
Debrecen	Virginia type	GI, M	TMV	TMV, AMV
Debrecen	Pallagi 3	M	TSWV	TSWV
Debrecen	Pallagi 1	M, Bli, Vn	TSWV	TSWV
Debrecen	Hevesi 17	Ldef, GI	TMV	PVY, CMV
Pallag	K-326	Mo, Olp	CMV	CMV
Pócspetri	Pallagi 3	Bli, Ldef, Chl	TSWV	TSWV

M: Mosaic, Mo, Mottle, Ldef: Leaf deformation, Vn: Vein necrosis, Chl: Chlorosis, Rs: Ringspot, Vc: Vein clearing, GI: Dark green islands, Bli: Blistering, Olp: Oak leaf pattern

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Summary

SURVEY OF VIRUS DISEASES OF TOBACCO FIELDS ON THE EASTERN HUNGARY

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Survey of virus diseases of tobacco fields on eastern Hungary was estimated by visual method, and the liability of visual inspection was controlled by ELISA serological tests. In 2003 the general infestation was relatively low, because of the dry and hot spring and summer, which did not favour for virus vectors. Among viruses tested TSWV was the most abundant one, followed by PVY, CMV, TMV and AMV. Visual characterization of TSWV was the most liable; but mosaic symptoms were due to more different viruses, such as TSWV, PVY, CMV and TMV. It is concluded, that visual characterization of virus diseases is very important, but not sufficient for testing plant viruses.

THE EFFECT OF BLACK SEED OIL FROM *NIGELLA SATIVA* AGAINST BARLEY POWDERY MILDEW DISEASE CAUSED BY *BLUMERIA GRAMINIS* F.SP. *HORDEI*

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Black cumin (*Nigella sativa* L.) is an annual dicotyledon of the Ranuculaceae family, growing in countries bordering the Mediterranean Sea. It has been employed for thousands of years as a food preservative and as a medicinal plant (Agarwal et al., 1979, Rahman et al., 2001). In Egypt, it is one of the widely distributed native plants (Farid et al., 2000). The seeds of black cumin are used in folk (herbal) medicine all over the world for the treatment and prevention of a number of diseases and conditions that include asthma, diarrhoea and dyslipidaemia (Ali and Blunden, 2003). The seeds contain both fixed and essential oils, proteins, alkaloids and saponin. Much of the biological activity of the seeds has been shown to be due to thymoquinone, the major component of the essential oil, which is also present in the fixed oil (Ali and Blunden, 2003).

It has been shown that the *N. sativa* oil exhibited strong antimicrobial activity against phytopathogenic fungi *Pythium vexans, Rhizoctonia solani* and *Colletotrichum capsici* (Rathee at al., 1982). Furthermore, the antifungal activity of black seed oil (BSO) extracted from *N. sativa* against *Rhizoctonia solani, Botrytis fabae, Fusarium solani, Alternaria tenuis* and *Sclerotinia sclerotiorum* was investigated by Rahhal (1997). He found that the essential oil of *N. sativa* was effective against all tested fungi except *B. fabae*. Farid et al. have demonstrated that BSO can control squash powdery mildew disease caused by *Erysiphe cichoracearum* (Farid et al., 2000).

Here we provide evidences that BSO can suppress the powdery mildew fungus of barley, (*Blumeria graminis* f.sp. *hordei*). To understand the mechanisms behind the effect against powdery mildew, we conducted *in vivo* histochemical staining for hydrogen peroxide, and also we analysed the spore germination and fungal growth after treatment with BSO using a microscope.

Materials and methods

Plant hosts and pathogens

Powdery mildew-sensitive barley (*Hordeum vulgare* L. cv. Botond and GK-Omega), tomato (*Lycpersicum esculentum* L. cv. Kecskeméti 549) and

cucumber (*Cucumis sativus* L. cv. Budai csemege) seeds were sown into soil and grown in greenhouse. Temperature was 18-23 ^{o}C, with 16 hours photoperiod per day using supplemental light with a light intensity of 160 µE m^{-2} s^{-1} and relative humidity 75-80%.
Tomato powdery mildew pathogen *Oidium neolycopersici,* cucumber powdery mildew *Podoshaera xanthii* and barley powdery mildew *Blumeria graminis* f.sp. *hordei* race A6 were maintained under greenhouse conditions and were used for all inoculation experiments.

Preparation of the oil
Black seed oil (BSO) from *Nigella sativa* was obtained from trade market in Egypt. Solution is ready for the spray or treatment contained 2 ml BSO and 1 ml Tween 20 in 1 litre. At first, the components and 4 ml water were shaking for 2-5 minutes and then we completed the volume to obtain one litre during shaking again (Farid et al. 2000).

The treaments of the plants with BSO
We treated three hosts (barley, tomato and cucumber) with the oil solution by spraying the plants twice 4-6 days after the inoculation with powdery mildew fungus to see the effect of the oil on different hosts. Barley, tomato and cucumber plants were 10-12 days, 8-10 weeks and 4-6 weeks old, respectively.
For microscopy investigations, we treated barley (cv. Botond) with BSO two hours before the inoculation and two days after the inoculation with the powdery mildew fungus.

Toxicity test
To determine the fungitoxic activity of BSO we used a cellophane water agar media (15 g agar and 1000 ml distilled water). Before pouring the agar medium in Petri dishes, 5 ml/l chloramphinicol antibiotic (1 % in ethanol) was added to the media. A cellophane membrane, well washed in boiling distilled water, was placed on the surface of water agar (Chet et al., 1981). We distributed the spores with sterilised brush on the surface of the cellophane and then sprayed the spores gently with BSO solution. Control Petri dishes contained only spores without oil treatment. After 18-24 hours, the rate of spore germination was tested by using Olympus BX 51 (Germany) microscope.

Preparation of samples for the microscopy
Leaves were harvested from control and BSO-treated plants one, two and three days after the inoculation. The detached leaves were immersed in clearing solution (0.15 % trichloroacetic acid in ethanol: chloroform 4:1) for

24 hours (Hückelhoven at al., 1999). Samples were kept in 50% glycerol before the microscopical examination. For the microscopy, the samples were stained with blue ink (Pelikan) for one minute, washed in water and put on glass slides.

Histochemical analysis of H_2O_2

Leaves were infiltrated with 0.1% 3,3-diaminobenzidine (DAB) in 10 mM Tris buffer (pH 7.8) for histochemical detection of H_2O_2. Samples were incubated under daylight for two hours after the vacuum infiltration. After the staining, leaves were cleared as described above and the intensity of brown colour was estimated by Chemilmager 4000 videodensitometer.

Results

The effect of BSO on powdery mildew disease

Powdery mildew disease was strongly inhibited in different hosts, such as tomato, cucumber and barley after treatment with BSO 4-6 days postinoculation. Under the influence of BSO, leaves of susceptible plants exhibited necrotic symptoms (Figure 1)

Figure 1. Effect of black seed oil (BSO) on the symptoms of barley (A), cucumber (B) and tomato (C) plants inoculated with powdery mildew. Untreated plants (left) and BSO-treated plants (right) were photographed at identical time points after inoculation

The inhibitory effect of BSO on spore germination

Rate of spore germination was determined on the artificial cellophane media. Table 1 shows the percentage of germinated conidia of powdery mildew fungi after treatment with BSO. Oil from *Nigella sativa* strongly inhibited the development of spores (Table 1).

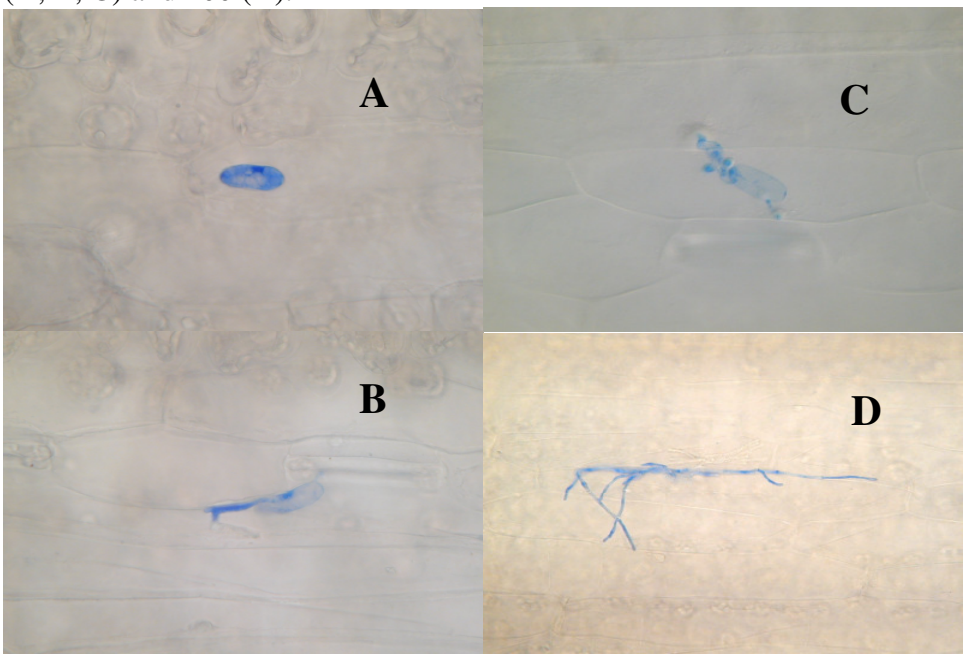
Table 1. Rate of spore germination of powdery mildew fungi on cellophane treated with black seed oil 20 min after inoculation. Results were evaluated 1 day after treatment

	Blumeria graminis f. sp. *hordei*		*Oidium neolycopersici*		*Podoshaera xanthii*	
	Control	BSO	Control	BSO	Control	BSO
% of spore germination	52	0.97	57	0.4	55	0.5
	58	0.00	60	0.00	59	1.4
	48	1.4	47	0.51	47	1.5
Mean	52.7	0.79	54.7	0.3	53.7	1.1
SD	5.0	0.7	6.8	0.3	6.1	0.6

Microscopic examination of the effect of BSO on the powdery mildew pathogen in barley

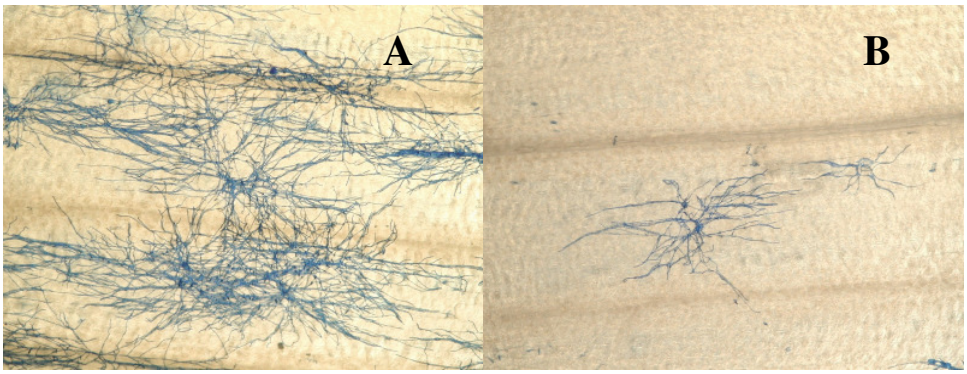
Spore germination was inhibited on barley leaves as well when we treated the plants with BSO 2 hours before inoculation (Figure 2A). The germinated conidia could not penetrate successfully the plant cell walls (Figure 2B)

Figure 2**.** *Blumeria graminis* f. sp. *hordei* on barley treated with black seed oil 2 hours before inoculation (A, B). Control leaves were untreated (C, D). Samples were harvested 24 h (A, C) and 48 h (B, D) postinoculation and visualized using an Olympus BX 51 microscope with a magnification of 800 (A, B, C) and 400 (D).



Growth of fungal mycelium was also inhibited by BSO when we treated the powdery mildew infected leaves 2 days after inoculation (Figure 3).

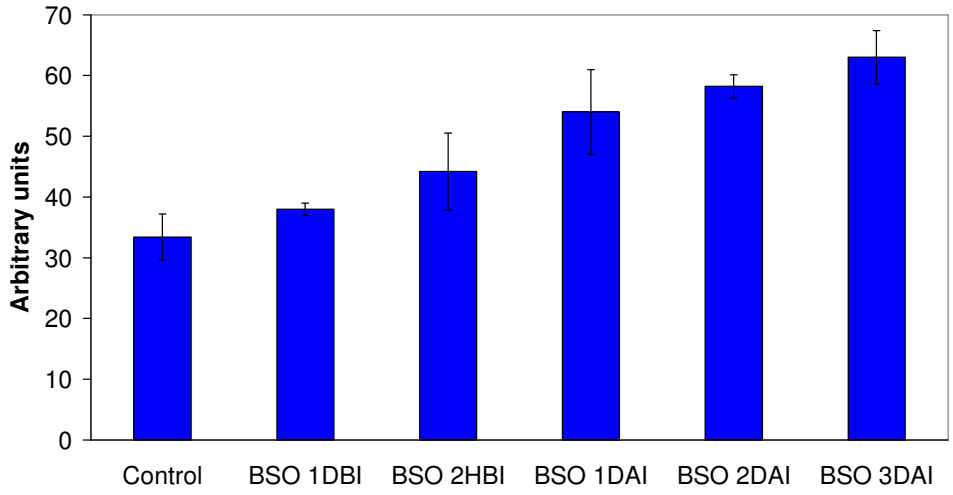
Figure 3. *Blumeria graminis* f. sp. *hordei* on barley treated with black seed oil 2 days after inoculation (B). Control leaves were untreated (A). Samples were harvested 3 days after inoculation and visualized using an Olympus BX 51 microscope with a magnification of 100x



Hydrogen peroxide accumulation

Hydrogen peroxide levels were detected in powdery mildew inoculated barley leaves using DAB staining. We have found that the BSO treatment increased significantly the level of H_2O_2 (Figure 4).

Figure 4. Level of hydrogen peroxide in barley leaves 4 days after inoculation with *Blumeria graminis* f. sp. *hordei*.



Plants were treated with black seed oil at different time points indicated on the figure. DBI: day before inoculation; HBI: hours before inoculation; DAI: days after inoculation. DAB staining was estimated by Chemilmager 4000 videodensitometer.

Discussion

Antimicrobial activity of black seed oil from *Nigella sativa* has been demonstrated by several researchers (Agarwal et al., 1979, Salem, et al. 2000, Ali and Blunden, 2003) related to human diseases. BSO can also protect plants from pathogens on arable lands (Rathee at al. 1982, Rahhal 1997, Farid et al. 2000). BSO was very effective against the powdery mildew fungus in squash (Farid et al., 2000). The present study indicates that susceptible barley, cucumber and tomato can be protected from the powdery mildew by treatment with BSO. The oil of seeds from *Nigella sativa* has strong toxic effect against the hyphal growth and spore germination. Conidia spores were damaged as a result of BSO treatment and development of germ tubes was inhibited. Interestingly, BSO not only exerted toxicity against spore germination but also increased the level of hydrogen peroxide (H_2O_2), which plays key role in the development of hypersensitive reaction (Hückelhoven et al., 1999). Therefore, BSO can

induce resistance response in the plant cells as well. In summary, BSO can strongly protect the plants against the powdery mildew pathogen by two distinct mechanisms, namely direct fungitoxicity and induction of plant resistance. We can recommend to give more attension to BSO for integrated pest management.

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PRELIMINARY RESULTS ON THE RESISTANCE OF TAN SPOT (*DRECHSLERA TRITICI-REPENTIS*) IN WHEAT AND DURUM VARIETIES AND CANDIDATES

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Tan spot (*Drechslera tritici-repentis*) is a very important disease in the western regions of the EU. The early drying of the leaves may cause serious yield losses. There are several reports about its significance in Hungary. The damage of the fungus depends on the weather conditions, the tillage system, the cultivation system and on the cultivated variety. Wet conditions, with periodic rainfall, reduced tillage and the loss of crop rotation aggravate the infection. The causal fungus does not like drought and extraordinarily high air temperature. So this year was not advantegous for DTR. The resistance of the varieties in the EU is usually well known, but in Hungary there were no official data about DTR resistance until now. There might be wet years in the future. In these years, tan spot may be as important as, the other well-known pathogens, like powdery mildew. The knowledge of the resistance is important for the producers.

Literature

Tan spot (*Drechslera tritici-repentis*) is a worldwide frequent pathogen, which has spread to the warmer regions since the 80's (5.). The fungus attacks wheat, durum, and some monocotyledonous weeds (Aponyiné-Garamvölgyi *et al.,* 1999; Obst and Gehring, 2002; Wolf, 1998). Damages caused by the fungus may be serious as reported by several authors. The average yield damage is at about 3 – 15 % (Hosford *et al*., 1987), but it might reach 50% (Hosford *et al*., 1987) also. In Germany, the damage might reach 30 % on susceptible varieties (Obst and Gehring, 2002). The pathogen likes the warm temperature up to 30^{o}C (Hosford *et al*., 1987), but dry conditions and too high temperatures reduce the pathogen's attack (Obst and Gehring, 2002). The causal fungus prefers the periodic long wet conditions (Hosford *et al*., 1987) and reduced tillage, which results in a lot of straw on the soil surface (Aponyiné-Garamvölgyi *et al.,* 1999; Bartels, 1999; Pringas *et al*., 2003; Wolf, 1998). The cultivation system without crop rotation

(Aponyiné-Garamvölgyi *et al.,* 1999; Obst and Gehring, 2002) is responsible for the stronger spreading too.
The fungus overwinters as mycelia on the straw, which can develop pseudothecia next spring (Obst and Gehring, 2002), or as mycelia in the infected seeds (Obst and Gehring, 2002; Schilder and Bergstrom, 1995). Seed infection may be significant, but seed dressing usually prevents the pathogen attack (Schilder and Bergstrom, 1995). The first infection materials are the ascospores, grown in perithecia in the early spring on the straw (Obst and Gehring, 2002). The ascospores cannot spread to a distance greater than some centimeters (Obst and Gehring, 2002; Sone *et* al., 1994). The secondary infection bases are the conidia, grown on the wall of the pseudothecia (Obst and Gehring, 2002), or on the infected leaf surface (Sone *et* al., 1994). The conidial spreading is very effective and the high temperature via the rising air may result in a wider spreading (Sone *et* al., 1994). The fungus causes symptoms of small brown-black spots with or without a yellow ring (Lamari and Bernier, 1989; Obst and Gehring, 2002; Strelkov *et al*., 2002). The presence and the size of the yellow ring based on the resistance of the variety (Lamari and Bernier, 1989; Strelkov *et al*., 2002). There is a great difference between the symptoms and the degree of damage of the resistant and susceptible varieties (Strelkov *et al*., 2002). The fungus has some divided races, which depend on the reactions of some varieties (Lamari and Bernier, 1989; Strelkov *et al*., 2002). About the Hungarian registered varieties there were no official data, the susceptibility of GK Öthalom published after the state estimation in 1999 (Aponyiné-Garamvölgyi *et al.,* 1999) was not justified in our trial.

Materials and methods

The provocative trial has been conducted at the Röjtökmuzsaj Experimental Station since 1999. The in vivo trial was performed in 2 replications, the registered varieties (in the last year) and the candidates were separated. The infection method was applying infected oat kernels and straw. On the oat kernels we applied 4 isolates from Germany (BBA-Braunschweig 4,8,12,12a), on the straw a Hungarian isolate from Szeged was used. There was a very virulent isolate at Röjtökmuzsaj, which appeared continuously from the first year on other wheat fields despite the crop rotation and the fungicide spraying. The oat was applied during the winter; the straw was applied at the end of autumn, when the plants were min. 5-6 cm high. Each plot was of 1 m^2 area, and irrigated the last time in May. Ploughing was not performed in the cultivation system. The amount of infective material was cca. 100 oat kernels and cca. 100 gramm straw at each plot. In the first 4 years, the infection was not strong enough, but it increased each year. On 10

June 2003, the leaf symptoms reached a measurable degree. We used a 5 grade scale, the minimum step was 0.25, which was equal to 5.0 % infected area on leaves. In the provocative trial we used all of the registered varieties and candidates, which were present in the official trials of Control of Value and Use. Wheat and durum varieties and candidates were present too. The total number of the variety candidates, varieties and durum varieties was 119, 109 and 5, respectively. Since the presence of other pathogens, primarily powdery mildew, may prevent the spreading of tan spot, the trial plots were sprayed during April and May 4 times with Calixin. During the second half of the vegetation period the pathogen was effectively isolated from the trial plots and the Septoria test made by Syngenta used as a control was always negative. Because the symptoms of tan spot may be similar to symptoms caused by other fungi, we separated and stored 5 typical infected leaves from each plot. These samples are free for controlling the dominance of the DTR in the trial plots.

Results

The first symptoms were observed on the trial field in the middle of April. The high air temperature and the draught prevented tan spot from fast spreading on the leaves. At the beginning of June, the foliar infection reached measurable degree. In the group of the registered wheat varieties, the trial mean was similar in the first two groups, a significant drop was only detectable in the mid-late group (Table 1). The distribution in the three resistance groups more or less fits the normal distribution (Table 2). In the group of the candidates, the early group, the first two trials resulted similar means and distributions. The mid-early candidates had a significant differential means in the trials (Table 1), with a one-sided distribution to the advantage of the susceptible group (Table 2). The mid late candidates resulted a significant lower mean (Table 1) and a normal dispersion (Table 2). The distance between the most resistant and the most susceptible genotypes is great enough in all groups. There is no significant difference in the values of the most resistant and susceptible varieties between the registered varieties and the candidates, or durum (Table 1).

Table 1. Extremities in the resistance to tan spot (*Drechslera tritici-repentis*) in several maturity groups of the provocative trial

Code of the trial	Maturity group	Resistant genotypes	Leaf infection %	Susceptible genotypes	Leaf infection %
Registered varieties: M 1	Early	GK Csongrád Mv Tamara **Mean:**	7,5 10,0 **30,9**	Pobeda Mv Dalma	57,5 55,0
M 2	Mid-early	Boszanova GK Zugoly st. **Mean:**	12,5 15,0 **31,0**	Buzogány GK Mura	57,5 50,0
M 3	Mid-late	GK Holló st. Capo **Mean :**	10,0 12,5 **22,5**	Ludwig Gaspard	37,5 35,0
Candidates : I/A-1	Early	Tina GK Pántlika **Mean:**	7,5 10,0 **28,4**	GK Tiszatáj st. Mv 04-02	55,0 45,0
I/A-B	Early	GK Csillag GK Fülemüle **Mean :**	10,0 12,5 **30,8**	FK 01480 Gloria	47,5 45,0
II/A-1	Mid-early	GK Zala GK Zugoly st. **Mean :**	22,5 27,5 **37,9**	Academie HP 83-00	65,0 52,5
II/A-2	Mid-early	Vasvári-4 GK Jupiter **Mean :**	10,0 12,5 **20,8**	GK Huba KT-2-02	37,5 32,5
III/A-1	Mid-late	Winnetou GK Holló st. **Mean:**	5,0 7,5 **14,9**	GK Market Mv Magdaléna st.	25,0 25,0
Durum		Prowidur **Mean:**	12,5 **23,5**	Superdur	42,5

Table 2. Distribution in percentage of the genotypes in the trials based on resistance to tan spot (*Drechslera tritici-repentis*)

Code of the trial	Maturity group	**Resistant** (less, than 50 % of the mean)	**Intermediate** (between 50 and 150% of the mean)	**Susceptible** (more, than 150% of the mean)
Registered varieties: M 1	Early	20	65	15
M 2	Mid-early	12	76	12
M 3	Mid-late	17	66	17
Candidates: I/A-1	Early	16	68	16
I/A-B	Early	10	74	16
II/A-1	Mid-early	0	96	4
II/A-2	Mid-early	3	79	17
III/A-1	Mid-late	22	56	22

Discussion

The trials which contain candidates in their 1st year have more susceptible genotypes to tan spot (I/A-b;II/A-2). The later years' selection pressure usually ends this phenomenon . They might give lower yield, or quality due to the higher tan spot damage. There is a high chance for genetic plant protection in the case of tan spot, the difference between the resistant and susceptible varieties is large. This trial did not verify the previously reported susceptibility of the old variety GK Öthalom. This variety was present in each trial as a maturity time standard and it was not the most susceptible variety in any of the trials. On the basis of the experiment, GK Öthalom seems to be a variety with average susceptibility. There might be a problem with the mid-early candidates. The infection of the resistant ones was too high and there were too much susceptible genotypes in the trials. These results are based on one year only, and continuing the trials is absolutely necessary. The wheat producers need this information and there might be wet years, when this pathogen and the resistance against it would be more important, than in this extraordinarily dry and hot year.

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Summary

PRELIMINARY RESULTS ON THE RESISTANCE OF TAN SPOT (*DRECHSLERA TRITICI-REPENTIS*) IN WHEAT AND DURUM VARIETIES AND CANDIDATES

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Tan spot (*Drechslera tritici-repentis*) is an important fungal disease all over the world, where the external conditions are advantageous for its infection. The fungus prefers wet conditions, reduced tillage and cultivation without crop rotation. In the Röjtökmuzsaj Experimental Station 5 years ago the phytopathological department of OMMI has begun a provocative trial to clear up the resistance of the wheat and durum varieties to tan spot. The layout was in 2 replications in whole random in the candidates and the registered varieties too. The plot size was 1 m^2, and the territory was cultivated without crop rotation. The pathogen attack was aggravated by placing out artificially infected oat kernels and naturally infected straw. The assessment was made with a 5 grade scale, each minimal step was 0,25, which was equal to 5%. The earlier ripening groups had a little higher infected plant surface, than the mid-late varieties. The distance between the extremities (5 % 65 %)is great enough to refer to the possibilities of genetic protection. Because there are only one-year results, the trial must be continued.

RESULTS OF A TWO-YEAR STUDY ON INCIDENCE OF *BARLEY YELLOW DWARF VIRUSES, CEREAL YELLOW DWARF VIRUS-RPV* AND *WHEAT DWARF VIRUS* IN TURKEY

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Barley yellow dwarf viruses (BYDV-MAV, BYDV-PAV, BYDV-RMV, BYDV-SGV), *Cereal yellow dwarf virus-RPV* (CYDV-RPV) and *Wheat dwarf virus* (WDV) are the most important virus pathogens of cereal crops in the world.

BYDVs were first identified in Europe, in The Netherlands by Oswald and Houston (1951) and were confirmed in the United Kingdom by Watson and Mulligan (1957).

In North America, cereal diseases characterized by yellowing and stunting were observed former than in Europe. Widespread outbreaks with significant yield losses that probably caused by BYDVs occurred in 1907 and 1949 (Hewings and Eastman 1995).

BYDVs belong to the family of *Luteoviridae*. They are not mechanically transmissible, are phloem-limited in host plants, and are transmitted in a persistent manner by numerous aphid species. BYDVs have a very wide host range in the *Poaceae.* Beside cereals, BYDVs are often present in maize, rice and grasses. Maize is widely grown in many areas. Thus, maize can be a primary source of inoculum for cereals. BYDVs are very common in perennial grasses being also principal source in areas of northern and western Europe (Lindsten 1964; Plumb 1977). Most grasses are tolerant and do not show definitive symptoms of BYDV (Clark and Christensen 1972; Holmes, 1985). They are also dangerous from the point of wiev of epidemiology of BYDV.

In Turkey, Bremer and Roatikainen (1975) reported for the first time about cereal virus diseases transmitting aphids and leaf-hoppers. In spite of the fact that resistant varieties were grown in Turkey, the yield was drastical reduced in some varieties in many years.
Considering the taxonomy of viruses, CYDV-RPV belongs to the genus *Polerovirus* within the family *Luteoviridae*. Pocsai et al.(2002) found that the incidence of CYDV-RPV in cereal species was variable during 1994-2001 in Hungary. During this period the incidence of CYDV-RPV was the highest in 1999.
WDV was first described in the former Czechoslovakia by Vacke (1961). Yield reduction caused by WDV varied from 5% to 97% depending on the time of infection. The occurrence of WDV was first described in Hungary by Bisztray et al. (1988) in winter wheat and Pocsai et al. (1991) in winter barley. The economic importance of WDV in Hungary is more important than that of BYDV (Pocsai, 2001; Pocsai et al. l997a, 1997b, 1998a, 1998b, 1998c, 1999, 2002a, 2002b). WDV is a leaf-hopper-transmitted geminivirus occurring on barley, wheat, durum wheat, triticale and rye. The species *Psammotettix alienus* plays the main role in the transmission of this virus disease. A large population of this vector is frequently present on cereal-growing areas of Hungary. WDV is a frequently occurring virus disease in the European countries (Vacke, 1961, 1962, 1988; Lindsten et al., 1970; Tomenius and Oxelfelt, 1981; Bisztray et al., 1988, 1989; Lindsten and Vacke, 1988, 1991; Lindsten, 1991; Pocsai et al., 1991, 1998; Bendahmane et al., 1995; Szunics et al., 1997, 2000, 2002; Bakardjeva and Habekuss, 1998; Huth, 1998; Commandeur and Huth, 1998; Lindsten and Lindsten, 1993, 1998, 1999; Huth and Lesemann, 1994).
BYDV and WDV cause similar symptoms (stunting and leaf yellowing). The most characteristic symptoms on cereals in the field are dwarfing and increased tillering. Stunting may be so severe that heads fail to emerge. Stunted growth is not limited to above-ground parts; root formation is also inhibited in BYDV- and WDV-infected plants.
The aim of the present study was to determine the incidence of BYDVs, CYDV-RPV and WDV in the Turkish cereal growing areas.

Materials and Methods

In 2002 and 2003, a survey was conducted in Turkey for the determination of the incidence rates of *Barley yellow dwarf viruses*, (BYDV-MAV, BYDV-PAV, BYDV-RMV and BYDV-SGV), *Cereal yellow dwarf virus-RPV* (CYDV-RPV) and *Wheat dwarf virus* (WDV) in winter wheat, winter and spring barley fields at different locations of Turkey. In 2002, 540 winter

wheat, 140 winter and spring barley samples were collected at fifteen locations (Çanakkale, Balikeşir, Izmir, Afyon,
Nevşehir, Konya, Kayseri, Sivas, Tokat, Amasya, Çorum, Ankara, Eskişehir, Kirklaleri and Edirne). Ten leaf samples exhibiting the mentioned symptoms were collected from each field. In 2003, 300 winter wheat, 220 winter and spring barley and 20 durum what samples were tested. Samples were collected at four locations of Turkey (Edirne, Eskişehir, Konya and Kütahya). In 2003, twenty leaf samples were collected from each field for virus test.

The cereal samples were assayed by DAS-ELISA for the presence of *Barley yellow dwarf viruses*, *Cereal yellow dwarf virus-RPV* and *Wheat dwarf virus*. The leaf samples collected were homogenized using a leaf pressing machine with the addition of ELISA sample buffer solution at a ratio of 1:10. The diagnostic materials used for BYDVs and CYDV-RPV were made by Agdia Inc., while that used for the WDV were prodeuced by Sanofi Pasteur Phyto-Diagnostics (France). Serological reactions were evaluated using a Labsystems Multiscan Plus photometer at 405 nm.

Results

The results of ELISA-tests on the samples of winter wheat collected in Turkey in 2002 are shown in Table 1. BYDVs were present in twenty-four of the fifty-four tested fields of winter wheat.

The extent of infection with BYDVs ranged from 3.3 to 20% in fields of winter wheat showing dwarfing and yellowing symptoms. Among the BYDVs occurring in the infected samples, the BYDV-PAV was the most prevalent, followed by BYDV-RMV.

CYDV-RPV was present in seven fields of winter wheat. Its incidence varied from 5.71 to 40%. WDV was determined in eleven of fifty-four tested fields with an infection ratio in the tested samples between 3.3 and 40 %. No infection was found in Amasya or Çorum. BYDV-PAV was the only virus detected in Edirne and WDV was the only virus detected in Ankara. Neither CYDV-RPV nor WDV was determined in Tokat, Amasya, Çorum, Kırklareli or Edirne provinces.

Table 1. Results of tests on winter wheat samples collected in Turkey in 2002

Location and date of sample collection (1)		No. of tested fields (2)	No. of tested samples (3)	Results of the tests (4)						No. of infected plants (5)
				BYDV-PAV	BYDV-MAV	BYDV-SGV	BYDV-RMV	CYDV-RPV	WDV	
Çanakkale	April 24	3	30	1	0	0	0	1	0	1
Balikesir	April 24	1	10	0	0	0	2	4	0	4
Izmir	April 24	2	20	0	0	0	2	3	0	5
Afyon	April 26	7	70	4	0	0	2	3	7	13
Nevşehir	April 27	1	10	0	0	0	1	0	2	3
Konya	April 27	3	30	2	1	0	0	0	3	6
Kayseri	April 28	3	30	1	1	0	0	0	2	4
Sivas	April 28-29	7	70	6	1	0	1	3	4	13
Tokat	April 29	3	30	4	0	0	1	0	0	4
Amasya	April 30	1	10	0	0	0	0	0	0	0
Çorum	April 30	5	50	0	0	0	0	0	0	0
Ankara	April 30-May 1	4	40	0	0	0	0	0	3	3
Eskişehir	May 1-2	6	60	0	0	0	3	4	5	11
Kirklareli	May 5	6	60	3	0	0	1	0	0	4
Edirne	May 5	2	20	1	0	0	0	0	0	1
Total (5)		54	540	22	2	0	13	18	26	72

The ELISA results obtained for the samples of winter and spring barley collected in Turkey in 2002 are summarized in Table 2. BYDVs were present in seven of the fifteen tested fields of winter and spring barley. The extent of infection with BYDVs varied from 10 to 30% in fields showing leaf yellowing symptoms. Two BYDVs (BYDV-PAV, BYDV-RMV) were present. It is obvious from the data that BYDV-PAV was the most frequent virus. The results obtained here suggest that BYDV- PAV and BYDV-RMV

Table 2. Results of tests on winter and spring barley samples collected in Turkey in 2002

Location and date of sample collection (1)		No. of tested fields (2)	No. of tested samples (3)	Results of the tests (4)						No. of infected plants (5)
				BYDV-PAV	BYDV-MAV	BYDV-SGV	BYDV-RMV	CYDV-RPV	WDV	
Çanakkale	April 24	3	30	1	0	0	1	0	1	3
Konya	April 27	2	20	2	0	0	1	0	1	4
Tokat	April .29	5	50	2	0	0	0	0	0	2
Çorum	April 30	1	10	0	0	0	0	0	0	0
Eskişehir	May 1-2	2	20	0	0	0	0	2	5	6
Kirklareli	May 5	1	10	1	0	0	2	0	1	3
Total (5)		14	140	6	0	0	4	2	8	18

could be of some significance in BYDV epidemiology. In general, among the cereal species the incidence of BYDVs in barley used to be the highest. This year the extent of infection with BYDVs recorded for winter wheat has exceeded that of winter barley. Virus infections were found in five of the six barley-growing provinces tested, but no virus was detected in Çorum. CYDV-RPV occurred only in one field with an incidence of 20%. WDV occurred in five of fourteen barley fields tested with an incidence rate ranging from 3 to 25%. The highest degree of infection (25 %) was detected in Eskişehir.The results of ELISA-tests on the samples of winter wheat collected in Turkey in 2003 are summarized in Table 3. BYDVs were present in eleven of the fifteen tested fields of winter wheat.

Table 3. Results of tests on winter wheat samples collected in Turkey in 2003

Location and date of sample collection (1)		No. of tested filds (2)	No. of tested samples (3)	Results of the tests (4)						No. of infected plants (5)
				BYDV-PAV	BYDV-MAV	BYDV-SGV	BYDV-RMV	CYDV-RPV	WDV	
Edirne	May 5	0	0	0	0	0	0	0	0	0
Eskisehir	May 8-11	11	220	12	0	10	8	1	28	55
Konya	May 9	4	80	2	0	2	16	3	5	25
Kütahya	May 10	0	0	0	0	0	0	0	0	0
Total (5)		15	300	14	0	12	24	4	33	80

The incidence of BYDVs varied from 5 to 50% in fields of winter wheat showing dwarfing and leaf-yellowing symptoms. The degree of infection was especially high in the infected fields of Konya. Among the BYDVs occurring in the infected samples, the BYDV-RMV was present at the highest rate. CYDV-RPV occurred only in two fields of winter wheat. Its incidence varied from 5 to 15%. WDV was present in thirteen of fifteen tested fields and its incidence ranged from 5 to 35%.
The ELISA results obtained for the samples of winter and spring barley collected in Turkey in 2003 are shown in Table 4.

Table 4. Results of tests on winter and spring barley samples collected in Turkey in 2003

Location and date of sample collection (1)		No. of tested fields (2)	No. of tested samples (3)	Results of the tests (4)						No. of infected plants (5)
				BYDV-PAV	BYDV-MAV	BYDV-SGV	BYDV-RMV	CYDV-RPV	WDV	
Edirne	May 5	4	80	0	0	0	0	0	26	26
Eskisehir	May 8-11	2	40	6	0	2	3	0	13	21
Konya	May 9	2	40	1	0	3	1	0	1	5
Kütahya	May 10	2	40	0	0	1	5	4	1	8
Total (5)		10	200	7	0	6	9	4	41	60

BYDVs were present in five of the ten tested fields of winter and spring barley. The extent of infection with BYDVs varied from 5 to 45%. The highest infection (45%) was found in Eskişehir (Alpu). Three BYDVs (BYDV-PAV, BYDV-RMV, BYDV-SGV) were present. Among the BYDVs, BYDV-RMV was the dominant virus. BYDV infections were found in three locations but there was no any infection of BYDV in Edirne. CYDV-RPV was present in two fields at Kütahya and its incidence varied from 5 to 15%. WDV was detected in seven of ten barley fields tested and its incidence rate ranging from 5 to 80%. The highest degree of infection (80%) was detected in Edirne.
The results of ELISA-tests on the samples of durum wheat collected in Turkey in 2003 are shown in Table 5.

Table 5. Results of tests on durum wheat samples collected in Turkey in 2003

Location and date of sample collection (1)		No. of tested fields (2)	No. of tested samples (3)	Results of the tests (4)						No. of infected plants (5)
				BYDV-PAV	BYDV-MAV	BYDV-SGV	BYDV-RMV	CYDV-RPV	WDV	
Konya	May 9	1	20	1	0	1	2	0	1	4
Total (5)		1	20	1	0	1	2	0	1	4

BYDVs (BYDV-PAV, BYDV-RMV and BYDV-SGV) were present with an incidence of 15 %. CYDV-RPV did not occur, and WDV was present in five percent.

Discussions

From the data of the serological tests, it is obvious that the incidence rates of BYDV, CYDV-RPV and WDV changed depending on the locations and cereal species. Little detailed information has been published on the incidence rates of BYDVs, CYDV-RPV and WDV in cereals, but it is evident that WDV must be expected to be a spreading and limiting factor in cereal production.

In general, the three main viruses of BYDVs (BYDV-PAV, BYDV-SGV, BYDV-RMV) and CYDV-RPV occur in Turkey, but their distribution is not uniform. Recently it has become evident that BYDV-RMV and BYDV-SGV have some significance in BYDV epidemiology.

During the two-year-period, the occurrence of BYDVs in winter wheat ranged from 3.3 to 50 %. The frequency of CYDV-RPV in winter wheat ranged from 5 to 40%, while the occurrence of WDV varied from 3.3 to 40%. The incidence of BYDVs in winter and spring barley varied from 5 to 45%. The incidence of CYDV-RPV ranged between 5 to 20% and the WDV infection in winter and spring barley varied from 5 to 80%. It is clear from these data that the importance of WDV and CYDV-RPV can be increased in cereals in Turkey. From the data of this paper, it is obvious, that the importance of WDV is incresing in cereal-producing regions of Turkey.

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Summary

RESULTS OF A TWO-YEAR STUDY ON INCIDENCE OF *BARLEY YELLOW DWARF VIRUSES, CEREAL YELLOW DWARF VIRUS-RPV* AND *WHEAT DWARF VIRUS* IN TURKEY

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A survey of cereal species was conducted in Turkey for the determination of the incidence of *Barley yellow dwarf viruses* (BYDV-MAV, BYDV-PAV, BYDV-RMV, BYDV-SGV), *Cereal yellow dwarf virus-RPV* (CYDV-RPV) and *Wheat dwarf virus* (WDV) during 2002-2003.

In 2002, 540 winter wheat, 140 winter and spring barley samples were collected at fifteen locations and in 2003, 300 winter wheat, 220 winter and spring barley and 20 durum wheat samples were tested. Samples were collected at four locations.The cereal samples were assayed by DAS-ELISA.

In 2002, the incidence of BYDVs in winter wheat ranged from 3.3 to 20% and in the samples of winter and spring barley varied from 10 to 30%. CYDV-RPV in winter wheat was detected in seven fields and the degree of infection varied from 5.71 to 40% and in winter and spring barley it was present only in one field with an incidence of 20%. WDV in winter wheat was present in eleven fields. Its extent of infection ranged from 3.3 to 40%. In winter and spring barley the degree of WDV infection varied from 3 to 25%.

In 2003, the incidence of BYDVs varied from 5 to 50% in winter wheat and from 5 to 45% in winter and spring barley. The incidence of CYDV-RPV ranged from 5 to 15% both in winter wheat and winter and spring barley samples.WDV infection varied from 5 to 35% in winter wheat while in winter and spring barley it ranged from 5 to 80%. From the data of this paper, it can be concluded that the importance of WDV is increasing in cereal-growing areas of Turkey.

DETECTING PEACH TREE SHORT LIFE (PTSL) DISEASE COMPLEX AT THE PLANT VARIETY TESTING STATION OF THE NATIONAL INSTITUTE FOR AGRICULTURAL QUALITY CONTROL IN TORDAS

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Three to six year-old trees are most susceptible to PTSL, although older or younger trees can also be affected. In the typical sequence, normal tree growth occurs for the first two to five years after planting. Wilt symptoms appear suddenly in the spring, and tree death often follows within a few weeks. During warm spring weather, bark on the trunk crotch, and scaffold limbs develop reddish, wet appearance, often accompanied by droplets of yellow or orange exudates. A "sour-soap" odour often is associated with tissues with this appearance. Further examination of the trunk and scaffold limbs may reveal cracks in the outer bark that usually extend inward to the xylem. In general, primary roots appear healthy, but feeder roots may be sparse, discoloured or necrotic. It is common for new shoots to grow from the base of the trunk by mid- or late summer (Ogawa et al., 1995).

PTSL is a complex disease, with numerous biotic and abiotic factors interacting. Causal agents can be described as those directly associated with tree death and those that predispose trees to the direct factors. Direct factors are freeze injury, bacterial canker (caused by *Pseudomonas syringae pv. Syringae* van Hall), and perhaps Leucostoma canker (caused by *Leucostoma cincta* (Fr.) Höhn.). Predisposing factors are the ring nematode (*Criconemella xenoplax* (Raski) Luc & Raski), rootstocks, time of pruning, root injury caused by cultural practices, and physical characteristics of the orchard site, such as pH and soil structure.

Trees pruned in late fall or early winter are more likely to succumb to PTSL than are trees pruned later. Cultural practices that damage tree roots, such as excessively deep cultivation, may also increase susceptibility to PTSL. Trees grown in acidic soils are more susceptible than trees grown in soils where the soil pH is maintained in the range of 6-6.5. Compacted soil or a hardpan may also inhibit tree growth and enhance susceptibility (Ritchie *et al*., 1982).

Materials and Methods

The infected trees died after bloom and leaf emerge in Tordas. The flowers and the young leaves dried on the twigs. All aerial parts of the trees were destroyed. In July new shoots came out from the base of the trunks. The

bark and the trunk of one of the killed trees cracked. I could saw voluminous orange exudates gurgling out of the holes of the shot-hole borer (*Scolytus sp*.). At the base of the trunks on the bark black pycnidia were visible on the dead trees.
I could smell the 'sour-soap' odour, when I cut out the trunks of the peach trees for further examination (16.07.2003). This special smell and the cracked bark plainly distinguished PTSL from peach tree replant problem.
The peach variety trial was planted in 1998. The area was arable land before planting. The plantation showed a normal growth the previous years. The dead and the dying trees were located randomly in the field. That's why any special soil spot or nematode infection did not interact as a predisposing factor. The disease incidence seemed to be independent from the variety. The disease severity (dead and dying trees) was 2.3% in the plantation.
Three trunks of dead peach trees were of variety 'Krümcsanyin'; 'Redhaven Bianka' and 'Pegaso' transferred to the Laboratory of Plant Protection Research Institute of Hungarian Academic of Science to identify the plague.
The identification process performed the morphological characteristics of the causal agent Leucostoma canker by László Vajna.

Results and Discussion

The PTSL complex disease was detected successfully in Tordas. The causal agent proved to be the Leucosoma canker *Leucostoma cincta* ((Fr.) Höhn.). Due to some environmental or cultural factors the young trees became stressed. The stressed trees attracted the shot-hole borers. These tiny bugs created entry points for the primary pathogen and served as vectors.
We can see a great descend of cultural practices in peach growing in Hungary. The lack of irrigation and proper fertilising, omitting deep ploughing before planting and the increasing pollution lead to stressed trees. That's why we are going to face an increase in the incidence of peach tree short life disease.

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Summary

DETECTING PEACH TREE SHORT LIFE (PTSL) DISEASE COMPLEX AT THE PLANT VARIETY TESTING STATION OF THE NATIONAL INSTITUTE FOR AGRICULTURAL QUALITY CONTROL IN TORDAS

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Peach tree short life (PTSL) is a disease complex. It can be clearly characterized by a sudden wilt of newly emerged flowers and leaves of peach trees in their third to sixth year after planting. It causes the death of branches or the entire aerial portion of the tree. Usually, apparently healthy trees may turn immediately to dead or dying trees. In the south-eastern United States, several million peach trees have been killed by PTSL during the past 30 years. At one of the infected sites tree mortality exceeded 50% in one year (Ogawa, *et al*., 1995). We observed the symptoms of PTSL at the Plant Variety Testing Station of the National Institute for Agricultural Quality Control in Tordas.

LECTURES OF MYCOLOGICAL SESSION

AN ALTERNATIVE PATHWAY OF GALACTOSE CATABOLISM IN *ASPERGILLUS NIDULANS*

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D-galactose is an important component of the plant cell wall sugar polymers known as hemicellulose. Its metabolism via the Leloir pathway is a ubiquitous trait in pro- and eukaryotic cells (Frey 1996). It can be used as a catabolic pathway for the degradation of D-galactose as an energy and carbon source while it links as an anabolic pathway the metabolism of carbohydrates, such as the synthesis of lipopolysaccharides, of cell wall components, and of exopolysaccharides. In cucumber, Leloir pathway was reported to be involved in the sucrose synthesis from stachyose (Gross and Pharr 1982). The Leloir pathway represents in the absence of an external D-galactose source the only means to provide this carbohydrate by biological interconversion from D-glucose to D-galactose. It involves an ATP-dependent galactokinase (EC 2.7.1.6) to form D-galactose 1-phosphate, which is subsequently transferred to UDP-glucose in exchange with D-glucose 1-phosphate by D-galactose 1-phosphate-uridyltransferase (EC 2.7.7.12). The resulting UDP-galactose is a substrate for the reaction catalyzed by UDP-galactose 4-epimerase (EC 5.1.3.2), resulting in UDP-glucose.

While the Leloir pathway appears to be the main D-galactose degrading pathway, alternative pathways of galactose catabolism have been reported in bacteria. D-galactose that has been transported via a galactose-specific phosphotransferase system (Gal-PTS) and been released into the cytoplasm as galactose-6-P is catabolized via the D-tagatose 6-phosphate pathway (Bettenbrock and Alpert 1998). In addition, an oxidative catabolism via 2-keto-3-deoxy-6-phosphogalactonate in bacteria has also been reported (Shuster and Doudoroff 1967).

Yeast loss-of-function mutants in galactokinase are unable to use D-galactose as a carbon source. However, in the filamentous fungus *Aspergillus nidulans* mutants in the *galE* (galactokinase-encoding) gene could grow on D-galactose in the presence of ammonium – but not nitrate ions – as nitrogen source. Mycelia of the wild-type strain of *A. nidulans* accumulated some intracellular galactitol (50 mM), whereas the *galE* mutant accumulated a 10-fold higher intracellular concentration. The accumulated galactitol was catabolized lateron in both strains. An *A. nidulans* loss-of-function mutant in arabitol-dehydrogenase, however, was unable to catabolize the accumulated galactitol. Further, an *A. nidulans* mutant in hexokinase (*frA1*) was unable to grow on galactitol, and a *galE / frA1* double mutant was unable to grow on either galactose or galactitol. Mycelia of *A. nidulans frA1*, pregrown on glycerol and transferred into a galactitol-containing medium, accumulated intracellular L-sorbose, indicating that the product of galactitol oxidation is L-sorbose. L-sorbose is a substrate for hexokinase, demonstrated by a loss of L-sorbose phosphorylating activity in an *A. nidulans* hexokinase (*frA1*) mutant. L-sorbose catabolism involves a hexokinase step, evidenced by the inability of the *frA1* mutant to grow on galactitol or L-sorbose, and by the fact that a *galE/frA1* double mutant of *A. nidulans* was unable to grow on D-galactose. The results therefore provide evidence for an alternative pathway of D-galactose catabolism in *A. nidulans*, which involves the NADPH-dependent reduction of the D-galactose to galactitol and NAD^+-dependent oxidation of galactitol by L-arabitol dehydrogenase to L-sorbose. An interesting feature of this novel pathway is that it does not involve pathway-specific enzymes, but makes use of proteins which have already been shown to be involved in other pathways.

Since the fungal aldose reductase is active with D-galactose (Singh and Schügerl 1992), it is likely that this enzyme is involved in the reductive pathway of D-galactose catabolism. Unfortunately, no *A. nidulans* mutants are known which are impaired in aldose reductase. This may be due to the fact that there are at least two, but likely more, aldose reductases present in filamentous fungi (Hasper et al. 2000; see also at www-genome.wi.mit.edu and microbial.cereon.com). If our hypothesis on the involvement of the aldose reductase in galactitol formation is correct, then this could be explained by the requirement of aldose reductase for NADPH, which may not be available during growth on nitrate because of the requirement and high affinity of the nitrate and nitrite reductases (Bhushan et al. 2002).

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Summary

AN ALTERNATIVE PATHWAY OF GALACTOSE CATABOLISM IN *ASPERGILLUS NIDULANS*

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D-galactose is metabolised via the Leloir pathway in pro- and eukaryotic cells. In the filamentous fungus *Aspergillus nidulans*, an alternative pathway of D-galactose catabolism was described. It involves the NADPH-dependent reduction of the D-galactose to galactitol and NAD^+-dependent oxidation of galactitol by L-arabitol dehydrogenase to L-sorbose. An interesting feature of this novel pathway is that it does not involve pathway-specific enzymes, but makes use of proteins which have already been shown to be involved in other pathways.

Role of Hydrogen Peroxide in Symptom Expression and in Plant Immunization

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Role of hydrogen peroxide in symptom expression

Three genotypes of barley (cultivar Ingrid) expressing the genes *Mlo* (susceptible), *Mla12* (resistant with HR symptoms) and *mlo5* (resistant without HR) in relation to infection by race A6 of *Blumeria graminis* f. sp. *hordei* have been sprayed with a solution of 20-50 mM H_2O_2 after establishment of infection (2-3 days after inoculation). Under the influence of H_2O_2, leaves of susceptible *Mlo* and *mlo5*-resistant plants exhibited HR-type symptoms with tissue necroses (Figs. 1 and 2).

The *Mla12*-resistant genotype produced HR earlier and the number of necrotic lesions increased, as compared to untreated control leaves (Fig. 3). It was possible to reverse the inhibitory as well as the HR-producing actions of H_2O_2 with injection of leaves with a combination of 2500 units/ml superoxide dismutase (SOD) and/or 5000 units/ml catalase (CAT) before treatment with H_2O_2.

It is suggested that the hypothetical negative regulation of HR-associated resistance in susceptible plants carrying the gene *Mlo* as well as in barley displaying HR-independent resistance and carrying the gene *mlo5*, could be associated with the limited production of H_2O_2 in infected plants. This action of H_2O_2 is sensitive to antioxidant enzymes, such as SOD and CAT (Figures 1-3).

Figure 1. Induction of HR in a susceptible combination of barley cultivar Ingrid (*Mlo*) with race A6 of powdery mildew. (A): Infected leaves with powdery mildew symptoms. (B): Uninfected leaves treated with 50 mM H_2O_2. (C): Infected and H_2O_2-treated leaves with symptoms of HR. Treatment was applied after establishment of infection (2 days after inoculation). (D): Infected and H_2O_2-treated leaves injected with a mixture of SOD and CAT. Symptoms of HR were reversed to the original symptoms of susceptibility

Figure 2. Induction of HR in a resistant cultivar displaying HR-independent resistance (Ingrid carrying the gene *mlo5*). (A): Infected symptomless leaves. (B): Uninfected leaves treated with 50 mM H_2O_2. (C): Induced HR with the stimulated necrotization in infected and H_2O_2-treated leaves. Treatment was applied after establishment of infection (2 days after inoculation). (D): The stimulated HR was reversed in infected and H_2O_2-treated leaves which were injected with SOD + CAT

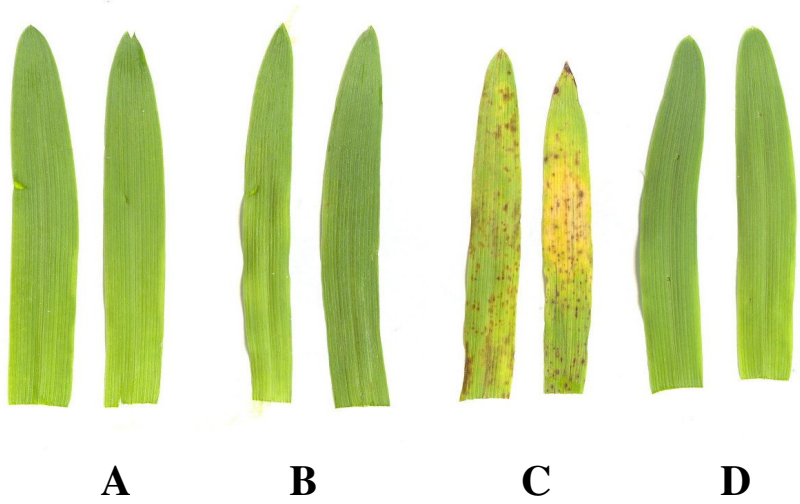


Figure 3. Stimulation of HR in the resistant cultivar Ingrid (*Mla12*) infected with race A6 of powdery mildew. (A): Infected leaves exhibiting HR. (B): Uninfected leaves treated with 20 mM H_2O_2. (C): Stimulated necrotization in infected and H_2O_2-treated leaves. Treatment was applied 2 days after inoculation (after establishment of infection). (D): Stimulation of HR was reversed in infected and H_2O_2-treated leaves which were injected with SOD + CAT.

Role of hydrogen peroxide in plant immunization

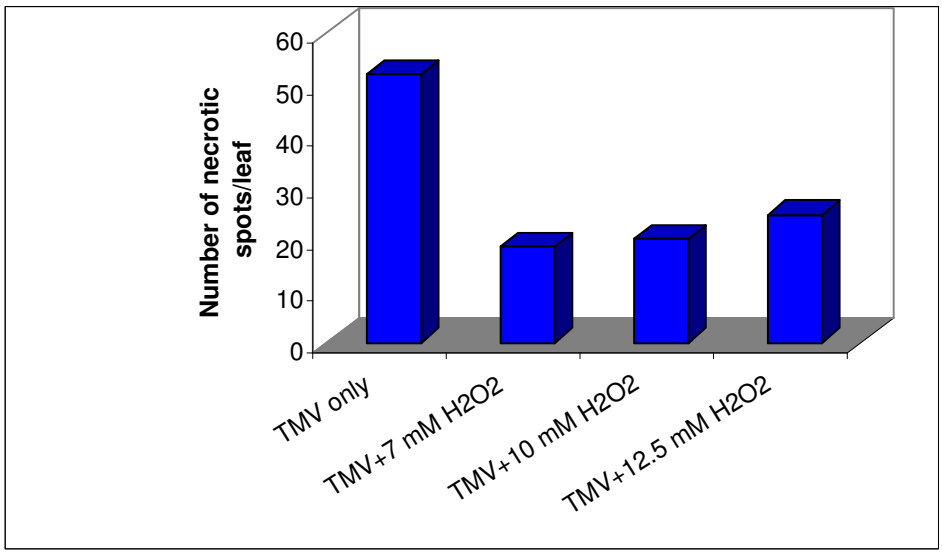
The research conducted in the Laboratory of Inzé (Gechev, T., Gadjev, I., Van Breusegem, F., Inzé, D., Dukiandjiev, S,. Toneva, V. and Minkov, I. (2002): Hydrogen peroxide protects tobacco from oxidative stress by inducing a set of antioxidant enzymes. CMLS, Cell. Mol. Life Sci. 59:708-714.) called the attention to the possibility to immunize plants which were sprayed with a low concentration of hydrogen peroxide (H_2O_2) against abiotic stresses. As was hypothesized, 5 mM H_2O_2 induces high antioxidant activities in treated leaves which resulted in suppression of damages caused by abiotic stresses.

We tried to immunize tobacco plants with 7-12.5 mM H_2O_2 against infection of tobacco mosaic virus (TMV) as well as to necroses caused by incompatible pathogenic bacteria. Figure 4 shows the reduced necrotization of the TMV-infected leaves which were sprayed with a water solution of 7 mM H_2O_2 one day before inoculation.

Figure 4. Effect of low concentration of H_2O_2 on necrotic symptom expression in TMV-infected resistant Xanthi-nc tobacco leaves. Left: Infected with TMV. Right: Leaf sprayed with 7 mM H_2O_2 one day before inoculation with TMV

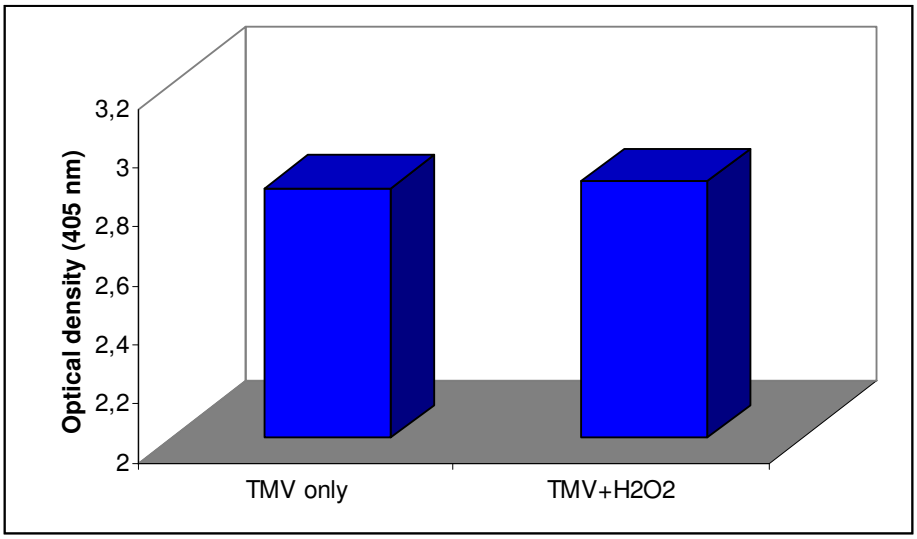
The reduction in the number of necrotic spots in the H_2O_2-treated leaves is shown in Figure 5.

Figure 5. Reduction of the number of TMV induced necrotic lesions, as a result of treatment of leaves with different concentrations (7-12.5 mM) of H_2O_2 one day before inoculation with TMV



Interestingly enough, the concentration of the virus in leaves, which were treated with H_2O_2 did not change, as compared to the control. One can suppose that the lack of necroses can not influence the virus content, as determined by ELISA-test (Figure 6).

Figure 6. Concentration of TMV in virus-infected and H_2O_2-treated leaves of Xanthi-nc tobacco.

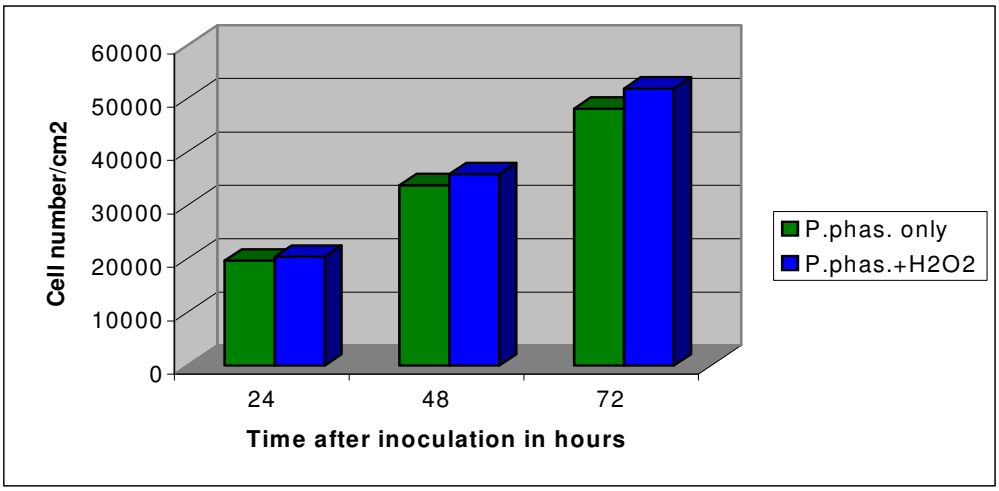


Immunization with H_2O_2 was also successful in relation to the HR-necroses caused by *Pseudomonas syringae* pv. *phaseolicola*. Spraying leaves with 10 mM H_2O_2 one day before inoculation almost fully suppressed the necrotic HR symptoms induced by 10^8/ml bacterial suspension (Figure 7).

Figure 7. Suppression of necrotization of Xanthi-nc tobacco leaves by treatment with H_2O_2 and inoculated with 10^8/ml bacterial suspension (*P.syringae* pv. *phaseolicola*). Left: Xanthi-nc leaf tissues injected with the bacterial suspension. Right: The whole leaf sprayed with 10 mM H_2O_2 and then tissues injected with the bacterial suspension one day after the spray.

In this case too, the number of bacteria in leaves which were treated with H_2O_2 did not change, as compared to the control. Consequently, the lack of necroses can not influence the multiplication of bacteria in leaf tissues (Figure 8).

Figure 8. Concentration of *Pseudomonas syringae* pv. *phaseolicola* in bacterially infected and H_2O_2-treated leaves of Xanthi-nc tobacco.



In summary, treatment of barley leaves with high concentrations of H_2O_2 induces HR-type necroses after inoculation with the powdery mildew fungus. On the other hand, the relatively low concenrations of H_2O_2 immunize tobacco leaves against tissue necrotization (HR) caused by TMV or *P. syringae* pv. *phaseolicola*. One can suppose that low concentrations of H_2O_2 stimulate the antioxidant capacity of the treated leaves. Antioxidants suppress oxidative burst (production of reactive oxygen species), thereby inhibiting the development of necrotic spots in infected leaves.

THE WEATHER COMPONENT OF APPLE SCAB EPIDEMICS IN ENVIRONEMTAL-FRIENDLY APPLE PRODUCTION SYSTEMS

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Venturia inaequalis (Cke.) G. Wint. is still one of the most destructive and economically important fungus in apple orchards, causing considerable losses every year. It has been known for a long time that there is a strong relationship between the weather parameters and the epidemic progression of a plant pathogen. However, various weather parameters play different roles in the disease development (Jeger and Butt, 1984). In the case of apple scab, temperature, leaf wetness and relative humidity are considered to be the most important factors affecting infection and epidemic progression of the disease. The relationship between weather parameters and development of apple scab was determined by Mills (1944) and Mills and LaPlante (1951). He summarized the relationships in a table called Mills' table. The Mills' table categorises infection based on duration of leaf wetness and mean daily temperature. The table also calculates a latent period at a given temperature.

Nowadays, scab control strategies are mainly based on the Mills' table or the revised Mills' table (MacHardy and Gadoury, 1989). However, it is proved that the Mills' criteria alone can not give sufficient information about the occurrence of infection (Sutton *et al*., 1976; Scheer, 1980; MacHardy, 1994; Trapman, 1994). Authors concluded that besides leaf wetness and temperature, relative humidity and other weather factors can also play an important role in the disease development. These authors related several weather parameters to disease development of plant pathogens. They found that relationships can be given in mathematical functions and they may be useful in disease warning.

The aim of this study was to analyse the relationships between weather parameters and disease increase of apple scab; and to compute mathematical functions for these relationships in integrated and organic apple productions.

Materials and Methods

Orchard site and plant material

The study was carried out in two experimental orchards. One was in the centre of the Netherlands at Randwijk (Fruit Research Station), and the

other was in Hungary at Debrecen-Pallag (University of Debrecen). In both countries, orchards consisted of an integrated field treated according to the IFP guidelines (Anonymous, 1995, 1998a) and an organic field treated according to organic production guidelines (Anonymous, 1997, 1998b). In both experimental orchards, tree spacing was 3 x 1.25 m and all trees were pruned to a spindle shape. All observations were made only on the scab susceptible cultivar 'Jonagold'.

Experimental layout and disease measures

In the Netherlands, both the integrated and the organic treated fields were divided into 5 blocks. Within each block, five trees were selected randomly for observation. In Hungary, 3 blocks, with 7 trees each, were present in both the integrated and the organic treated fields. Disease incidence and severity were assessed on a weekly basis in 1998 and 1999 at Randwijk and in 1999 and 2000 at Debrecen-Pallag. The proportion of shoots, leaves and fruits diseased (disease incidence) was calculated separately as the number of diseased shoots, leaves and fruits divided by the number of shoots, leaves and fruits selected. To quantify the percentage of the total disease on shoot, leaf and fruit (disease severity) severity scales were composed according to Croxall *et al*., (1952a, 1952b). Assessments were done from the beginning of May until mid-October.

The weather component

To evaluate the role of the weather component in the apple scab epidemic, first, the weekly disease increase was determined at a certain week (n). Weekly disease increase was used as a dependent variable. Weekly mean temperature, relative humidity, rainfall, Mills' infection periods and interaction between temperature and relative humidity were used as independent variables. Weekly disease increase was related to rainfall, relative humidity, Mills' infection period, temperature and interaction between temperature and relative humidity. Five different periods were used in the analyses: i) week (n-1), ii) week n(n-1), iii) week (n-2), iv) week (n-1)(n-2). Simple and multiple regression analyses were performed to relate disease increase to several weather parameters (Sutton *et al*., 1976). The aim was to find the best regression models, which explain the role of disease increase in relation to weather parameters in integrated and organic production systems. Selection criteria of a regression model were:

- coefficients with positive sign in single and multiple regressions;
- constants and coefficients with a reasonably small standard error;
- P-value < 0.1 for each partial coefficient;
- as high coefficient of determination as possible.

Linear regression analyses were performed with Genstat statistical program package.
Sources of weather data were: a) daily mean temperature (°C) recorded at 1 m above ground, b) rainfall (mm) recorded with a standard pluviometer, c) daily mean relative humidity – RH (%) recorded at 1.5 m above ground level. Mills infection periods in hours were calculated by METY 2.3 scab warning system in The Netherlands, and by the Mills' table in Hungary.

Results

For both countries, significant relationships between weather parameters and disease increases can be seen in Table 1 and 2. Fruit incidence and leaf incidence gave the best relationships between disease increase and weather parameters. Fruit incidence gave good relationships with weather data in the organic production system. Mills infection period (Mills), relative humidity (RH), temperature (Temp), interaction between relative humidity and temperature (Temp x RH) and their combinations gave good relationship with fruit incidence in week (*n*-2). In Hungary, the relative humidity played a smaller role in the disease development in the organic production system than in the Netherlands. In the integrated production system, good relationship was found between leaf incidence and weather parameters in both countries. Leaf incidence showed relationship with Mills' infection period, relative humidity and different combinations of Mills' infection period with relative humidity and temperature in week (*n*-2).

Table 1. Relationship between disease increase of apple scab and weather parameters in week (*n*-2) in integrated and organic apple production systems (Randwijk, 1998 and 1999)

Organic			
Disease measurements	Weather parameters	Coefficient sign	R^2
fruit incidence	Mills	+	0.22*[a]
fruit incidence	RH	+	0.25*
fruit incidence	Temp + RH	+' +'	0.22*
fruit incidence	Temp x RH	+	0.21*
fruit incidence	Temp x RH + Mills	+' +'	0.37*
fruit incidence	Temp x RH + RH	+' +'	0.35*
Integrated			
leaf incidence	RH	+	0.62**
leaf incidence	Mills	+	0.75***
leaf incidence	RH + Mills	+' +'	0.81***
leaf incidence	Temp x RH +Temp	+' +'	0.68**
leaf incidence	Temp x RH +Mills	+' +'	0.85**

[a] F-test = *** < 0.01, ** 0.01 - 0.05, * 0.05 - 0.1

Table 2. Relationship between disease increase of apple scab and weather parameters in week (*n*-2) in integrated and organic apple production systems (Debrecen-Pallag, 1999 and 2000)

Organic			
Disease measurements	Weather parameters	Coefficient sign	R^2
fruit incidence	Mills	+	0.34**[a]
fruit incidence	Temp + RH	+' +'	0.24*
fruit incidence	Temp x RH	+	0.28*
fruit incidence	Temp x RH + Mills	+' +'	0.39**
Integrated			
leaf incidence	RH	+	0.32*
leaf incidence	Mills	+	0.55**
leaf incidence	RH + Mills	+' +'	0.43**
leaf incidence	Temp x RH +Temp	+' +'	0.49*
leaf incidence	Temp x RH +Mills	+' +'	0.68***

[a] F-test = *** < 0.01, ** 0.01 - 0.05, * 0.05 - 0.1.

Moreover, shoot incidence and leaf incidence gave strong relationships with temperature in week *n*(*n*-1) in both countries (data not shown).

Discussion

In this study, we determined the relationship between disease increase of apple scab and weather parameters in four examination periods. Significant relationship was found for fruit incidence in the organic and for leaf incidence in the integrated production systems (Table 1 and 2). The strongest relationships were found in week (*n*-2), which occurs two weeks before symptoms appeared. The length of incubation periods for apple scab ranges from 10 to 15 days (Tomerlin and Jones, 1982). We can conclude that disease increase was closely related to almost all weather parameters in the first part of the incubation period. Results also demonstrated that later at the time of / *n*(*n*-1) / week, temperature played a more important role in the fungus development than the water parameters (relative humidity, rainfall, leaf wetness). Consequently, the spore dissemination, spore germination, penetration and development of the first subcuticular hyphae are significantly dependent on almost all weather parameters, but during the incubation period the most important weather parameter is the temperature.
Relatively high correlations were found when 2 or 3 single weather parameters were related to disease increase in multiple regression analyses (Table 1 and 2.). The best correlations were always related with Mills'

infection periods. These relationships demonstrate that interaction between weather parameters play a more important role in the disease increase than a single weather parameter. Therefore, the practically useful combination incorporates interaction between temperature and relative humidity and Mills' infection period in week (n-2). The suggested equations are: y_1 = 0.0004 x + 0.006 z - 0.05 for organic, and y_2 = 0.0002 x + 0.004 z – 0.07 for integrated, where y_1 = weekly increase of fruit incidence, y_2 = weekly increase of leaf incidence, x = interaction between temperature and relative humidity, and z = Mills' infection period in hours.

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THE WEATHER COMPONENT OF APPLE SCAB EPIDEMICS IN ENVIRONEMTALLY FRIENDLY APPLE PRODUCTION SYSTEMS

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Relationships between disease increase of apple scab and weather parameters were determined and mathematical functions were computed for integrated and organic apple productions in a three-year study in the Netherlands and Hungary. To evaluate the role of the weather component in apple scab epidemic, first, the weekly disease increase was determined at a certain week (*n*). Weekly disease increase was related to rainfall, relative humidity, Mills wetness period, temperature and interaction between temperature and relative humidity. Five different periods were used in the analyses: i) week (*n*-1), ii) week *n*(*n*-1), iii) week (*n*-2), iv) week (*n*-1)(*n*-2). In both countries, fruit incidence and leaf incidence gave the best relationships between disease increase and weather parameters in week (*n*-2) in organic and integrated production systems, respectively. Disease increase was closely related to almost all weather parameters in week (*n*-2). Results also demonstrated that later, in week *n*(*n*-1), temperature played a more important role in the fungus development than the water parameters (relative humidity, rainfall and leaf wetness). Consequently, the spore dissemination, spore germination, penetration and development of the first subcuticular hyphae are significantly dependent on almost all weather parameters, but during the incubation period the most important weather parameter is the temperature.

PCR-BASED DNA MARKER FOR IDENTIFICATION OF INTERSPECIFIC *PHYTOPHTHORA* HYBRIDS ATTACKING ALDER TREES

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Detection and identification of species in the pseudofungal (oomycetous) genus of *Phytophthora* by traditional, mainly morphologically based approaches often lead to difficulties. Difficulties may, among others, arise when morphological similarities among species occur as a result of occasional interspecific hybridizations. Such hybrid phytophthoras are even more unlikely to be identified by conventional methods. Conclusive proof of interspecific hybridizations among related *Phytophthora* species either in laboratory or in nature has been obtained by molecular tools, i.e. by the detection of DNA sequences of biparental origin (*cf.* Érsek, 2002).

Of special interest in present work is an unusual, previously unknown *Phytophthora* that caused severe mortality of alder trees (*Alnus* spp.) in southern Britain in the mid 1990s and then has occurred widely across Europe including Hungary (Brasier et al., 1995; Szabó et al., 2000). Recent molecular evidence has shown that the alder *Phytophthora* is a hybrid of two developmentally rather different and alder non-pathogenic species: *P. cambivora* and *P. fragariae* (Brasier et al., 1999). The pathogen comprises a range of heteroploid hybrids, the 'standard' and the 'variants'. The 'standard' type occurs more commonly, and is more aggressive than those collectively termed variants that are also present in several countries.

RAPD (**r**andom **a**mplified **p**olymorphic **D**NA) analysis of the Hungarian isolates of alder *Phytophthora* (the standard type and one of the variant types, *i.e.* the Swedish variant) revealed a well discernible, *ca.* 900-bp fragment of the standard type isolates, that was amplified by the 10-mer primer, OPG-02 (Nagy et al., 2003). This fragment did not occur in either of the parental species and a range of other *Phytophthora* spp. Consequently, the relevant RAPD fragment appeared to be unique to the standard type isolates.

Because of the shortcomings of single random primers, we attempted to derive two different primers for PCR (**p**olymerase **c**hain **r**eaction) with even greater specificity to avoid the occurrence of irrelevant fragments. The oligonucleotide primer pair was designed on the basis of nucleotide sequences obtained from both ends of the isolated and then cloned

subgenomic RAPD fragment of the standard alder phytophthoras (SAP). The two (17-mer and 18-mer) primers, both comprising the sequence of OPG-02 (underlined), were as follows:

SAP1: 5'– GGC ACT GAG GGT TCC TC – 3'
SAP2: 5'– GGC ACT GAG GTC TAG ATT –3'

At the optimal annealing temperature of 60 °C, the PCR produced the anticipated single fragment of *ca.* 900 bp in the standard type isolates only. That is, no discernible PCR product occurred in any of the tested isolates of the Swedish hybrid variant and 21 *Phytophthora* spp. including *P. cambivora* and *P. fragariae* as parental species of the hybrid alder phytophthoras.

Conclusively, this novel PCR marker provides a powerful tool for rapid identification of the aggressive standard type hybrid. Since the reaction requires no more than 1 ng of template DNA, the procedure could be applied to detecting the pathogen in the nursery-raised alder stock to avoid the use of infected plants for reforestation.

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STUDY OF THE MYCOFLORA OF BÁTORLIGET II

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Bátorliget" situated in Szabolcs-Szatmár-Bereg country, in the Eastern border region of Hungary. The strictly protected ancient bog carries the evolution, organising and structural change of the Hungarian Great Plain in its history and development.

János Tuzson directed the attention of the scientific world to the „Bátorliget" Ancient Bog in 1914. The discovering botanical and zoological investigations started on the region in 1928. Due to this work, on the nature conservation area – established in 1938 – the strict protection of the ancient bog was enlarged to 53 hectares.

The extremely richness of species of the flora in the „Bátorliget" Ancient Bog (Boros 1932, Soó 1934, Simon 1956, 1991) remained from the woody-boggy-bare age of the Great Plain, namely from the beech-period, but its birch bogs originated from the birch-pine age. Some species (*Trollius europaeus* L., *Angelica palustris* /Bess./ Hoffm.) have glacial genesis (Soó 1953).

Ubrizsy investigated the fungus world of the „Nyírség" since 1937 (Ubrizsy 1940, 1941, 1943, 1947). He observed mainly microfungi (56 species), but he also found some large fungi in the „Bátorliget" Ancient Bog. Till now the most detailed picture about the large fungus association of „Bátorliget" Ancient Bog, wich list contains 86 large fungus species, published by Ubrizsy in 1953. Lenti and Máté (1995, 1996) served later information about the mostly unrevealed fungus elements of „Bátorliget" Ancient Bog – according to kind advices of professor József Vörös – continuing in spirit of inheritance of academician Ubrizsy.

The object of some year work to study the species of the large fungi, the presence and quantitative relations, connections with the separated units of the vegetation (Standovár et al. 1991) on „Bátorliget" Ancient Bog (and later on other protected area in the region). The data, won by the investigation of large fungi, can give such information about the past and present conditions, moreover the nature conservation values of „Bátorliget" Ancient Bog, wich we cannot form a complete notion about the area without.

Materials and Methods

The bog is situated in the Central European flora area of the Holarctic flora empire, in the flora-region of *Pannonicum*. More exactly, it is positioned in the „Nyírség" flora-area (*Nyírségense*) of the flora-region of Great Plain (*Eupannonicum*). Under number of EOTR 710-233 and number of MTB: 8299.2 officially, it belongs to the „Hortobágy" National Park as a forestry.
„Bátorliget" is an unique region in our country, where sub-alpine type plant and animal species live, as remainders of the flora and fauna of some cold, glacial or post-glacial period. The survivers have been rendered by the micro-climatic relation. The cold subsoil water, moving near to the surface, not only moistens, but cools the soil, so the layer of air near to the surface also remains cold. The evaporation of the bog-water increases the vapour content of the air, and the wreath of the forest prevents sweeping of the fog, forming even in the early morning of summer, over the bogs (Soó 1953).

The fungus species has been examined at the skirts of the willow-bog (*Calamagrostio-Salicetum cinereae*), which appears as the first tree-association of the boggy succession-line. There are some smaller birch-tree group here, too.
We have not done surveying of the large fungi on the birch-bog spots of the area, moreover in the associations of the different water-content meadow types, highlands and reeds yet.
In 2002 from January to the end of December we have done surveying of large fungi 24 times in the area of „Bátorliget" bog. The species were determined on the fields, or in laboratory, later on we conserved them as dried materials. We noticed the producing place, primining, their probable mycorrhiza partnerships.

For identification and classification we use the publications of the following authors: Moser (1983), Jülich (1984), Zerova et al. (1972, 1979), Krieglsteiner (1991-1993), Kits van Waveran (1985), Breitenbach et Kränzlin (1981-1995), Moser et Jülich (1985-1996), Arnolds et al. (1995) respectively. Data are stored and evaluated by the „Pilzkartierung 2000" PC programme of the German Micological Society (Seilt 1991, Rimóczi 1994). Categories for fungus ranks were used by Ainsworth and Webster (Rimóczy 1995).

Results

During our surveys we registered 511 basidiomycetous large fungus species on the area of „Bátorliget" Ancient Bog in 2002. It means four times more

species than the formerly known ones from „Bátorliget" published by Ubrizsy's (1953) work.
The taxonomy division of the species listed below:
32 species in the 12 genera of *Phragmobasidiomycetidae*, 24 species in the 8 genera of *Gasteromycetes*, 130 species in the 53 genera of *Aphyllophorales*, and the greatest number: 325 species in the *Boletales-Agaricales-Russulales* orders.

Data relate to the taxon-reachness and great diversity of the large fungal world of the area, on the base of the one year regular surveying, however some of genera with many species (*Entoloma, Cortinarius, Lepiota, Coprinus, Clitocybe,* etc.) have not occured in expected number yet.
We have not found species from such genera as *Melanoleuca, Cystolepiota, Gymnopilus, Lyophyllum*. It is certain, that these genera, moreover the species of *Naucoria* genus – which are characteristics for wet areas – are present with many species on „Bátorliget" Ancient Bog.
During our surveying we could not take attention for such large fungus families of *Ascomycotina*, as *Helvellaceae, Pezizaceae, Humariaceae, Geoglossaceae, Helotiaceae* or *Sphaeriaceae*, however we often saw the species from these taxa. Observing them will be the task for the next years, and as a result we count for numerous species even in the first year.
There were 348 saprobe species among the observed fungi. After a few year monitoring it will be practical to over-evaluate this life categories, distributing for 6-7 sub-categories (eco-groups), according to Winterhoff (1993) who made a long-term examinations of the large fungus world of the grove-forests, along the Rhine.

Now according to Krieglsteiner (1993), the saprobe fungi are separated as living on a dead and/or mouldering bedding, where we could place 177 species. An another group is fungi living on the very mouldered substrates, the number were 135.
Some species (for example *Coprinus disseminatus, Marasmius rotula*) were found in both groups, because in some cases they could live on different substrates. It also happend, that the same fungus appeared on leaf or a piece of twig (for example the *Marasminellus ramealis, Tubaria furfuraceae*).
Most of the identified fungi were saprobes. It is based on the character of the area. In the stocks of the grove forest and sandy oaks, the production of dry fallen leaves and parched grass is powerful, the fallen tree-trunks – which can moulder untouchedly – the fallen twigs mean a changeable living area for the saprophagous and xilophagous fungi.

In the leafy forests, out of the typical species (*Auricularia mesenterica, Cerrena unicolor*, *Clitocybe incornata*, etc.), there were fungi characteristic for hard-wood groves, among the saprobes: *Ramicola*

haustellaris, Pluteus phlebophorus, Squamanita schreieri, etc. Arnolds et al. (1995) and Kreisel (1981) defined them as *Alno-Padion* species.
It is not accidental, because the plants of the hard-wood groves show mutual features with the willow groves (*Salicetalia*), and hornbeam forest (*Fagetalia*). It is ranged to the latter, as one of the association of *Ulmion* (Soó, 1995), or recently the *Alnion* (Borhidi and Kevey 1996).

The fungi which are listed as typical ones in the „Bátorliget" Hard-wood groves, are also characteristic of the mountanious fresh leafy forests indeed. For this reason no wonder, that we have found some fungi, which were typical for the beech wood here: *Marasmius cohaerens, Antrodiella hoehnerii, Micromphale foetidum* (which is a typical genus of *Carici-Fagetum* by Kreisel 1981) and *Micromphale brassicales*. The latter two formed an „aspect" in a huge mass in the grove forest and bare oaks, at the end of August. Just like the *Macrotyphula filiformis*, which gave such a top production in the middle of September.

It is interesting, that due to the cold climate of the bog, such saprobe genera of the *Fagion* or *Carpinion* associations, as the *Megacollybia platyphylla* or the *Oudemansiella musida* grows on the *Quercus robur* dry fallen leaves and parched grass, or trunk.
The *Ganoderma adspersum* and *Ganoderma resinaceum* appearing as parasite on the living trees are also a mutual characteristic genus of the *Fraxino-Ulmetum* and the mountanious fresh *Fagetalia* associations.
The *Squamanita schreieri* is also present as a typical species of the grove forests. It characterises not only the association but occasionally warming up of the growing place, because in Europe it is signed from the rare grove forests of the river valleys, which are able to relatively warm up (Kreisel 1981).

The growing area of „Bátorliget" was the second place in the country, as the mentioned *Psathyrella silvestris* had been found first in the willow-alder bog by Babos (1989). The *Psathyrella melanthina* is also a typical grove forest species. Typical *Salicetalia* species: *Cytidia salicina*, *Exidia repanda*, *Phellinus conchatus*.
After the first year observations it is not practical to supply the number of the saprobiont genera sperately for the grove forest, respectively for the oaks, because they concern for a very small area, moreover some parts of the testing areas belonged to the transition area between the two associations.

During the next studies we intend to examine properly the bounding features, frequency, distribution to the different substrate types and tree species.

The fifteen (15) obligatory parasite fungus genera – which we have monitored – fortunatelly are not so many. It gives a good notion about the state of health of the tree-stock on the area.
Most of these parasite tinders have a wide host spectrum, for example: *Ganoderma lucidum, Phellinus contiguus*. Most of them link to the *Quercus* genus (for example: *Inonotus dryadeus, Phellinus robustus*). The presentation of the tinders, which often sponge on *Fagus*, is not suprising on *Alnus* (*Inonotus obliguus*) or *Salix* (*Phellinus ferreus*).

The 148 pieces of mycorrhiza fungus mean near one-third of the genera. The twenty-three genera refer to very wide range of the large fungus taxonomy. There were not any obligatory pine-mycorrhiza fungus among the mycorrhiza genera, because there is not pine forest plantation on the area of „Bátorliget" Ancient Bog, the natural occurrence of the pine varieties can be left out of account. We have not done examinations around the scattered pine-trees. Although their fungus-connections could be interesting in this situation.
Generally, from the group of pine mycorrhiza fungi the most various tree-partners occurred with *Quercus robur* (*Xerocomus badius*) or *Betula pendula* (*Amanita muscaria*).

Russula faginea, associated to beech-tree, is a typical *Carici-Fagetum* species according to Einhellinger (1987) and Kreisel (1981), that was also a typical one in the hornbeams-oaks. Arnolds et al. (1995) and Galli (1996) mentioned it from oaks, too. Wee have also found under the *Quercus robur* on the „Bátorliget" Ancient Bog. Similarly, the *Hygrophorus chrysodon* has also been found, which is also a fungus of *Fagetalia*.

The eighty percent of the mycorrhiza genera associated with *Quercus robur*: 119 fungus genera. Eleven species are birch-tree mycorrhiza. We have also found some mycorrhiza fungi, associated with poplar and alder tree.

The fungi, associating with birch-tree, have been found mostly on fields of *Calamagrostio-Salicetum cinereae* and under the outside birch samples, but we have not surveyed on the residue birch-bog, which were signed in the vegetation map of the bog.

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STUDY OF THE MYCOFLORA OF BÁTORLIGET II

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„Bátorliget" Ancient Bog is situated in the Eastern border region of Hungary, which has been a nation-wide nature conservation area since 1950. It survived from the woody-boggy-bare age of the Hungarian Great Plain, but its birch bogs originated from the birch-pine period in all probability. Some species of plants and animals are glacial genesis.

The investigation of the fungal associations have been started in 1995, and after the discovering work of one year we could make a report about the basidiomycetous large fungi of „Bátorliget" Ancient Bog. We have determined 511 pieces of the basidiomycetous large fungus species in the half-natural and young stocks of the pedunculated oak-silver lingen-tree forest. They can be ranked into 138 genera of the 29 fungus family by identification survey.

The grove-forest fields are very poor in mycorrhiza species: just one-ten part of the mycorrhiza fungus lives here. Probably it is a natural condition for the biotop, because Winterhoff (1993) found the same rates among the saprobiont-mycorrhiza along the Rhine, and Krisai -Greilhuber (1992) along the Danube.

After a one year monitoring we have found some protecting fungus species on the area of „Bátorliget" Ancient Bog: *Amanita caesarea, Amanita muscaria, Amanita solitaria, Boletus regins, Boletus satanas, Boletus rhodoxanthus, Gyroporus cyanescens*, species of *Geastrum* genus, morover *Psathyrella melanthina*, and *Psathyrella sylvestris*.

EXAMINATION OF HUNGARIAN POPULATIONS OF *CRYPHONECTRIA PARASITICA* (MURR.) BARR ON OAK

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Cryphonectria parasitica (Murr.) Barr (syn: *Endothia parasitica* [Murr.] And.) causes big damages of the chestnut-stands throughout the world. First at the beginning of the 20th century killed almost the whole American chestnut (*Castanea dentata*) populations in the USA. After the pathogen was transferred to Europe in the middle of the 20th century and infected the European chestnut (*Castanea sativa*) populations in Western Europe and caused the disease named „chestnut blight".

This serious disease had spread gradually from the West-European chestnut sites towards the eastern territories and arrived to the Carpathian-Basin too. Its occurrence was observed first in Hungary in 1969(Körtvély, 1970), in Slovakia in 1976 (Juhasova, Radócz, 1999), in Romania in 1985 (Florea, Dumitrescu, 1999), and in Ukraine in 2001 (Radócz, 2001).

Nowadays the Middle- and East-European region are considered as the"frontline" of spreading of *C. parasitica* by experts.

Up to now this disease had been the most important disease for European chestnut. However the importance of the disease is still more increasing because it is able to infect other tree species of *Fagaceae* plant family (oak, beech). Therefore it can cause potentially serious damages in our forests. There are 21.6% fine oaks and 11.4% austrian oak (*Quercus cerris*) on the Hungarian forest territory. One of the most important varieties of the Hungarian forestry the sessile oak (*Quercus petrea*). These oak trees are important both economically and ecologically.

We had made our investigations in 2002 on Pécsbányatelep, Mecsek Mountain, Southern-Hungary in a chestnut-forests mixed with 9% oaks and 13.5 % beech trees.

The goals of our studies were the follows:

Estimating damages caused by *Cryphonectria parasitica;* laboratory investigation of the collected samples and isoletes; evaluation and analysis of the results achieved.

Materials and Methods

Time, place and standpoints of the sampling
We made our field examinations on 17-18 of May, 2002 on Pécsbányatelep in a mixed chestnut forest with 6.5 hectares territory (chestnut trees – 86.3%, beech trees – 13.5%, oak trees – 9%, others – 1.2%) to check the disease symptoms and infection rates. We examined all the oak trees and 100 randomly selected chestnut trees. We registered the number of the infected, killed and recovering trees and we collected bark samples for laboratory isolations and furthermore investigations.

Laboratory examinations
We put the surface sterilized bark samples to potato-dextrose-agar media (PDA). Incubation of samples lasted for 7 days in a climate chamber. After a week we transferred again developed mycelia of fungus to get pure cultures.
The vegetative compatibility tests were made by pairing the isolates with EU-tester strains (EU-1-31). The vegetatively compatible isolates groupped in the same VCG (Vegetative Compatibility Group). Those isolates which produced a barrage zone at the edge of the growing mycelia were classified into different VC-groups.
During laboratory processes we made virulence tests with isolates on pieces of branches from chestnut with 1 cm diameter and 25 cm of length. Chestnuts were inoculated by 4 isolates had been origined from chestnut and 4 ones from oak. After two weeks of incubation the length of developed necroses were measured.

Results and Discussion

Field examinations
We had examined 165 trees on the site including 100 chestnut and 65 oak trees.
The chestnut blight disease was spread through the territory and symptomes were observable well. However there were differences in the mortality of two tree species, 50.8% and 92% of the examined oak and chestnut trees were infested respectively. The number of dead trees were higher among the infected chestnuts (19%) than among the oaks one (9.2%).

Vegetative compatibility tests
We had collected 23 samples from infested or killed chestnut trees and determinde their compatibilities. We can see the results in the Table 1.

Table 1. Vegetative compatibility tests of PB 1-23 isolates from chestnuts

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23
1	x																						
2	+	x																					
3	+	+	x																				
4	-	-	-	x																			
5	+	+	+	-	x																		
6	-	-	-	+	-	x																	
7	+	+	+	-	+	-	x																
8	+	+	+	-	+	-	+	x															
9	-	-	-	+	-	+	-	-	x														
10	-	-	-	+	-	+	-	-	+	x													
11	+	+	+	-	+	-	+	+	-	-	x												
12	+	+	+	-	+	-	+	+	-	-	+	x											
13	+	+	+	-	+	-	+	+	-	-	+	+	x										
14	+	+	+	-	+	-	+	+	-	-	+	+	+	x									
15	-	-	-	+	-	+	-	-	+	+	-	-	-	-	x								
16	+	+	+	-	+	-	+	+	-	-	+	+	+	+	-	x							
17	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	x						
18	+	+	+	-	+	-	+	+	-	-	+	+	+	+	-	+	-	x					
19	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	+	-	x				
20	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	x			
21	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	x		
22	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	+	x	
23	+	+	+	-	+	-	+	+	-	-	+	+	+	+	-	+	-	+	-	-	-	-	x

Remarks: + vegetatively compatible isolates
- vegetatively incompatible isolates

We can see that there are more than one compatibility groups.
We had made this comparisons on oak isolates as well. 33 samples were collected from infected or killed trees. There were more than one compatibility groups similar to chestnut isolates.

Vegetative compatibility examinations
We paired the isolates with the EU-tester strains and determined their compatibility. Results with isolates from chestnuts are shown in the Table 2.

Table 2. Vegetative compatibility tests of PB- 1-23 isolates from chestnuts with EU 1-13 tester strains

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23
1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	+	+	+	-	+	-	+	+	-	-	+	+	+	+	-	+	-	+	-	-	-	-	+
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
8	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
9	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	-	-	-	+	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	+	+	-
12	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Remarks: + vegetatively compatible isolates
- vegetatively incompatible isolates

The chestnut isolates were compatible with the EU-3 and EU-11. But there were incompatibility with the other EU-tester strains. On the basis of the results we can conclude that the sexual multiplication of the fungus can be important on this growing area and the genetic variability of *C. parasitica* is significant. We examined the bark samples by microscope and we observed peritecia (teleomorph stage of the fungus).
We can come to similar conclusions with the oak isolates. These were compatible with 3 EU-testers (EU-3, EU-11, EU-12) as shown in the Table 3.

Results of the virulence tests
We had measured the necrosis of caused by each isoletes after two weeks of incubation. We established that all the examined strains of the fungus had caused cankers on the bark. The length and the width of the cankers were measured and calculated their infested area. On the basis of the results we can establish that the cankers were larger when the branches were infected isolates from oak. It can concluded that isolates from oak were more virulent than those isolates collected from chestnuts.

Table 3. Vegetative compatibility tests of PBT 1-33 isolates from oaks with EU- 1-13 tester strains

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33
1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3	-	+	-	+	+	-	-	+	-	-	-	-	+	-	-	-	-	+	-	-	-	+	-	+	+	-	-	+	-	-	-	-	+
4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
8	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
9	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
11	+	-	+	-	-	-	-	-	+	+	-	+	-	-	-	-	-	-	+	+	+	-	-	-	-	+	+	-	+	-	-	-	-
12	-	-	-	-	-	+	+	-	-	-	+	-	-	+	+	-	+	-	-	-	-	-	+	-	-	-	-	-	-	+	+	+	-
13	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Remarks: + vegetatively compatible isolates - vegetatively incompatible isolates

The importance of the disease on oaks

There were proved that the founded cankers on the trunk of the oak trees had been caused by *Cryphonectria parasitica* infection. Laboratory isolations and identifications confirmed the findings. However infested oak trees occurred only in mixed forest with chestnut. All the isolates were virulent, hypovirulent strains were not able to infect oaks (or perhaps it is possible but very rare).

On the basis of the results we can establish that *C. parasitica* have not caused so serious destruction on oaks as on chestnuts until now, but potentially it could be a serious disease of oak species in the Carpathian-basin.

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EXAMINATION OF HUNGARIAN POPULATIONS OF *CRYPHONECTRIA PARASITICA* (MURR.)BARR ON OAK

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Cryphonectria parasitica caused serious damages of the chestnut stands throughout the world in the last century. This disease appeared in Hungary and other East-European countries too. It could cause serious potential damages for our forests. Our chestnut trees are seriously infested and other important tree species can be damaged such as oak or beech causing signifacnt losses both economically and ecologically.

We investigated a chestnut forest mixed oak on Pécsbányatalap, South-Hungary. We eastablished that *C. parasitica* infected not only the chestnut but also the oak trees as well. Although the contamination and the caused damage was smaller on oaks than chestnuts, it could be more serious disease for oak species in the future.

LECTURES OF WEED SCIENCES & INTEGRATED PEST MANAGEMENT (IPM) SESSIONS

EFFECT OF PHENOLOGY AND RAINFALL ON ALLELOPATHY OF *XANTHIUM ITALICUM* MOR.

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The cocklebur species become more and more significant weed species in Hungary, where the row crops (such as: corn, sunflower, sugar beet, etc) are endangered by them very badly. Their fast spreading and danger are caused by many reasons: lasted sprouting, reduced sensitivity against many kinds of herbicides, competitional ability, allelopathy, fast initial growth and changes in climate.

Allelopathy is used by many species of plants in the competition with one other, as well as by the cockleburs. But the way and efficiency of this are influenced by many factors. Species with allelopathical effect could have different attitudes under different conditions: they can produce materials with retarding and stimulating effects in different quantities or composition, and the actual conditions of existences (water supply, nutritive supply, quantity and quality of light, proportion of minerals etc.) of the acceptor plants and the mediator agent (i.e. soil) influence the final effect.

Literature

Cocklebur species are spreading, noxious weeds, and get in the centre of interest in several parts of world. However, there are experiments for therapeutic utilization in some country, e. g. against *Plasmodium falciparum, Trypanosoma evansi* (Joshi et al. 1997; Talakal et al. 1995).

Ground cover of cockleburs, especially *Xanthium strumarium* and *X. italicum*, became larger and larger in the past decades in Hungary, like warm-philous species (Szőke, 2001). They threaten mainly row crops (sugarbeet, maize, sunflower, soybean) and cause reduce of yield (Bloomberg et al. 1982; Wilson, 1995).

Danger of cockleburs may be explained with continuous emergence, large capability for competition, allelopathy and spreading several pathogens, e. g. they are hosts of beet necrotic yellow vein virus (Dikova, 1997; Kutluk et al. 2000), in addition, plants are toxic at cotyledon stage when seeds contain hydroquinone (Mitch, 1987).

Allelopathy of *X. sturmarium* was examined on several cultivated plants: lettuce, maize, soybean (Inam et al. 1987), and extracts were active against *Proteus vulgaris, Staphylococcus aureus, Bacillus subtilis, Candida*

albicans and *C. pseudotropicalis* (Jawald et al. 1988), moreover, extracts of *X. italicum-pennsylvanicum* complex inhibited *Azotobacter, Nitrobacter, Rhizobium* strains (Rice, 1964).
The allelopathy of plants was caused by several compounds, among others free amino acids, phenols (Elmore, 1980a; Colton and Einhellig, 1980; Elmore, 1980b), which may have several other functions. Their role was studied exposed to stress factors where their level increased in tissues of several species preventing decay of certain plants. (Sircelj et al., 1999; Sanchez et al. 1998; Gilbert et al., 1998; Ashraf et al., 1994; Maggio et al., 2002; Politycka, 1999). Consequently, stress factors can effect on allelopathy, and modify interactions between plants. These allelochemicals can be produced in different ratios and quantities under different conditions (e.g. drought stress, different nutrient supply) modifying the interference between weeds and crops (Dávid and Radócz, 2002).

Materials and Methods

Xanthium italicum and *Abutilon theophrasti* Medic., from which extracts were made, were grown in the field with different population densities (5 plants per m^2 and 20 plants per m^2). Samples were collected in May at four or five leaves stage and in July (before flowering) before and after rainfall. There were long drought periods before 20 and 15 mm rain. Fresh roots and shoots were cut into small pieces, 4g of fresh biomass was put into 100 ml tap water and left for a day, then extracts were filtered through filter paper. Test plants were cress (*Lepidium sativum* L.) and sugarbeet (*Beta vulgaris var. saccharifera*). Experiments were conducted in Petri plates on filter paper with four replications at room temperature used 6ml leachate and 50 seeds of test plant in each dish. Root and shoot growth of cress were measured on the 3^{rd} and 6^{th} day, and germination of sugarbeet was valued on the 6^{th} and 10^{th} day.

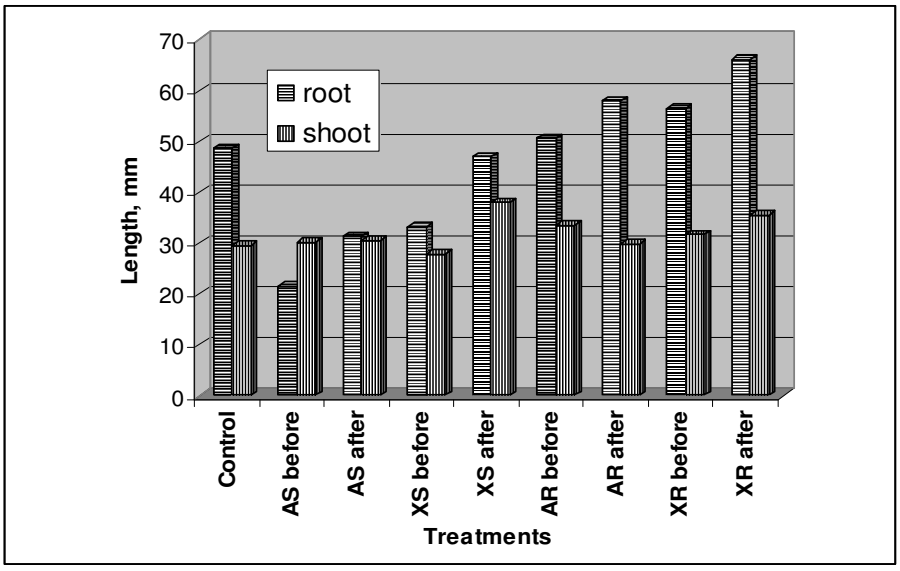
Results

Experiments were conducted in May and in July, in both cases samples collected before and after rainfall. There was difference between the effect of root and shoot extracts depending on the phenological stage and rainfall.

Effect on growth of cress in relation to rainfall
Leachates of shoots of cockleburs and velvetleafs collected before rainfall in May reduced root length. Leachates made from roots had no inhibitory effect, in fact, samples after rainfall slightly promoted the growth. (Samples

collected before rain had no significant effect compared to check.) The difference in shoot growth was smaller (Figure 1).

Figure 1. Effect of velvetleaf's and cocklebur's leachates on root and shoot growth of cress before and after rainfall in May; A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.



In July there was a larger difference between the effects of extracts made from shoots collected before and after rainfall than in May. Leachates before rainfall strongly retarded root growth, however, after it leachates had no inhibitory effect, in fact, leachate of shoots of low population density stimulated the growth. The effect was similar but slighter on shoot growth. Extracts made from roots had no significant effect on growth of cress except treatment with roots of high population density, which promoted root growth (Figure 2).

Effect on germination of sugarbeet in relation to rainfall

Germination of sugarbeet was examined by treatments with the same leachates. In May, every extract depressed the germination on the 6th day, but extracts made from plants collected after rainfall exerted stronger effect than those made from plants collected before rainfall. The difference was large in the case of cocklebur's extracts, and small in the case of velvetleaf's extracts (Figure 3). By the 10th day the difference became smaller for treatments with shoot extracts, but a significant difference was observed in the case of root extracts made before and after rainfall.

Figure 2. Effect of cocklebur's leachates on root and shoot growth of cress before and after rainfall in July; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain H = high population density.

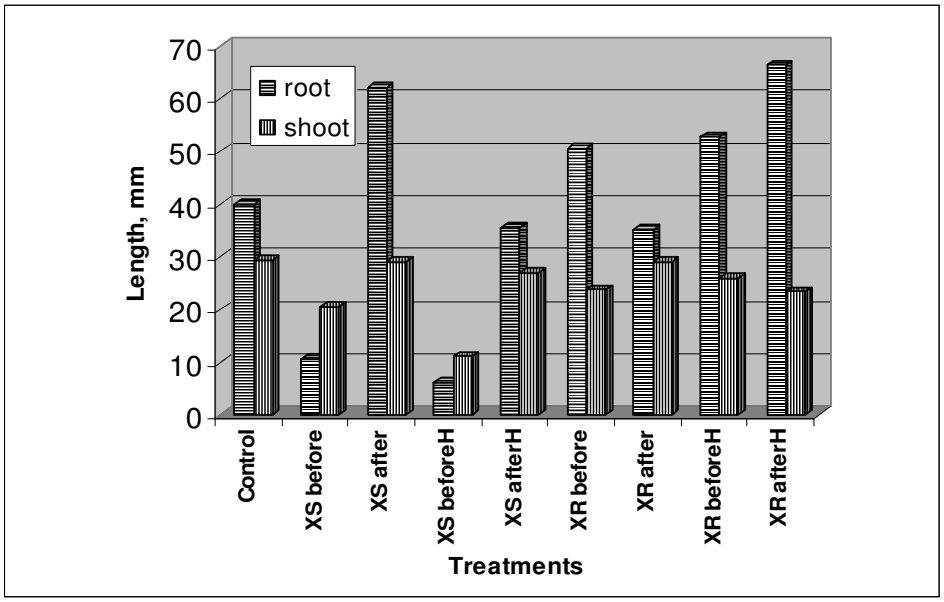
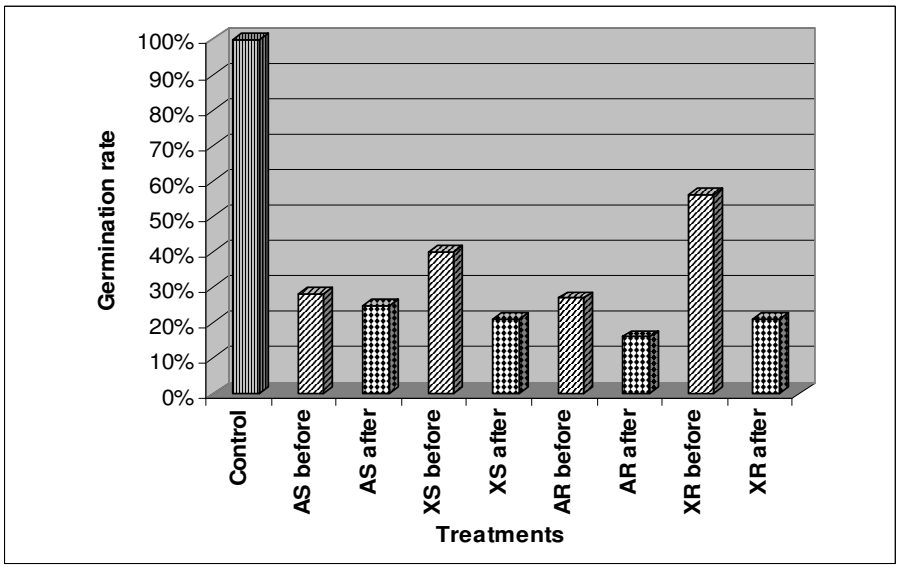
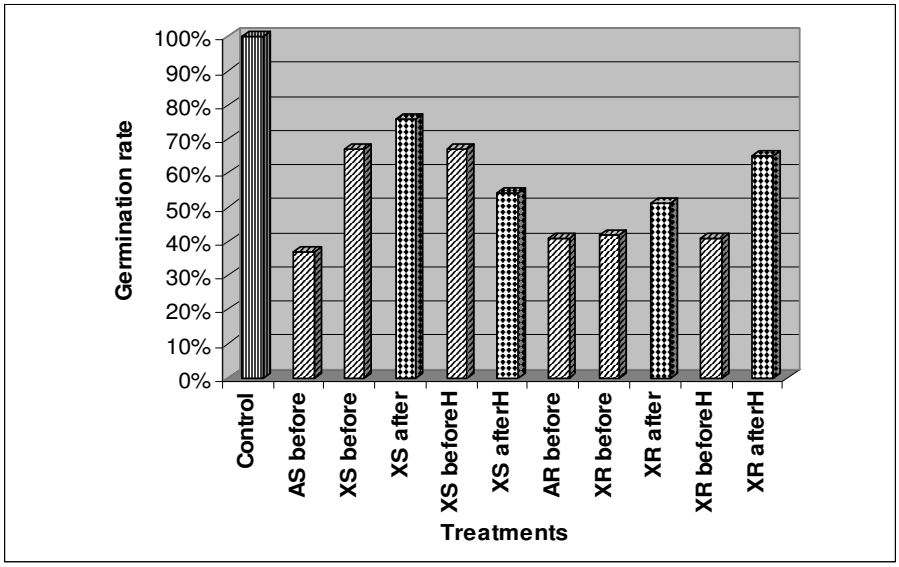


Figure 3. Effect of velvetleaf's and cocklebur's leachates on germination of sugarbeet on the 6th day before and after rainfall in May; A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.



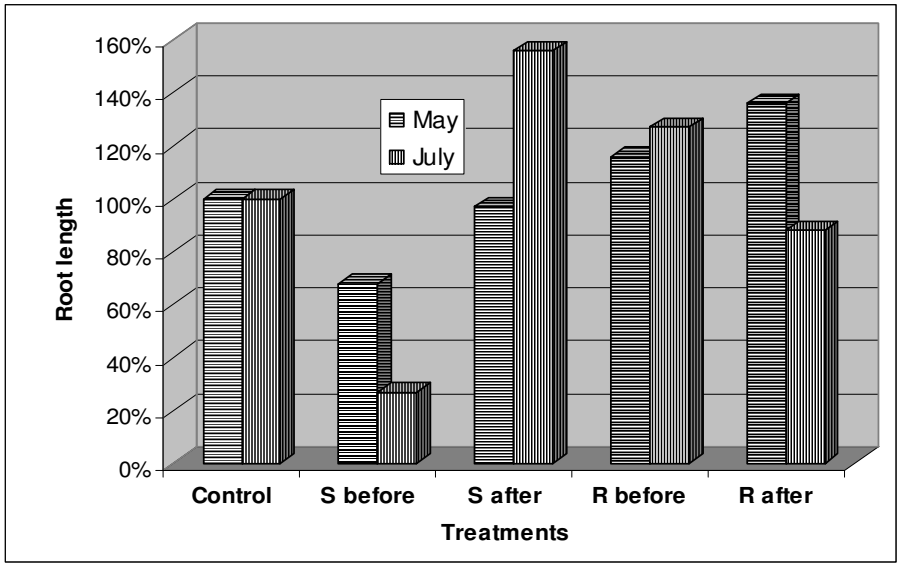
In July the inhibition of germination was mostly weaker, and the difference was smaller before and after rainfall than in May. The effect of rainfall was significant only in the case of cockleburs' root extracts of high density population (Figure 4).

Figure 4. Effect of velvetleaf's and cocklebur's leachates on germination of sugarbeet on the 6th day before and after rainfall in July;
A = leachate from velvetleaf; X = leachate from cocklebur, S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain, H = high population density.



Difference caused by phenology
Radicle length of cress was strongly retarded by shoot extract made before rain in July, but the effect was smaller in May. In the case of shoot extracts after rain, no effect was observed in May, and a promotive effect in July. Rainfall influenced effects of root extract differently in May and July (Figure 5).

Figure 5. Effect of cocklebur's leachates on root growth of cress in May and July; S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain.

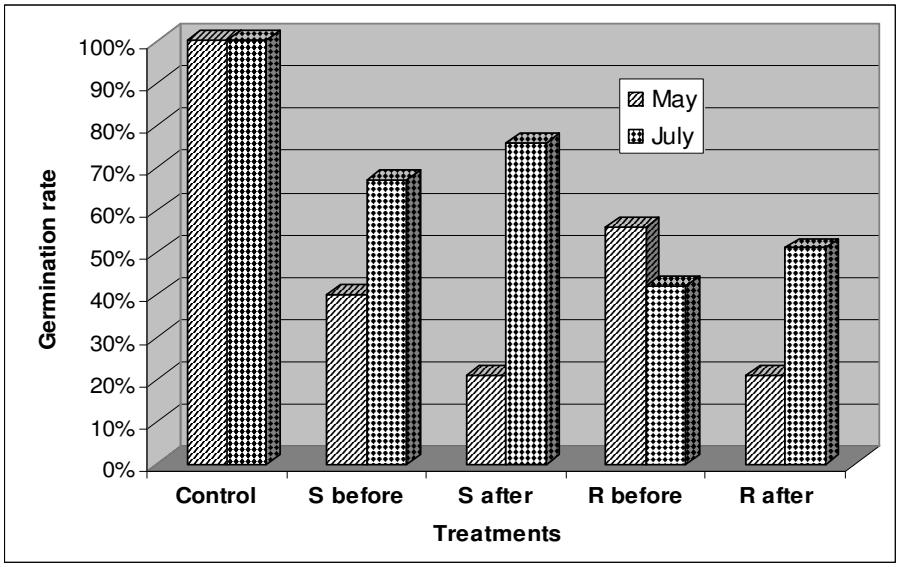


Trend was similar in the case of shoot growth, but only slight difference was observed between results in May and July.
On the 6th day, germination of sugarbeet was retarded more strongly in May than in July by shoot extracts, and the difference was especially large after rainfall. Root extracts before rain had a similar effect, but after rainfall the germination was significantly lower in May than in July (Figure 6). By 10th day the difference did not decrease in the case of root extracts.

Discussion

The results supported the hypothesis that some environmental factors and the phenological stage of plants play a determining role in interference between individual plants. Results can be altered depending on only the date of sample collection. When interpreting and comparing results of allelopathical experiments, it is recommended to pay attention to environmental conditions, phenological stage, setting of experiment etc.

Figure 6. Effect of cocklebur's leachates on germination of sugarbeet on the 6th day before and after rainfall in May and July
S = shoot extract, R = root extract, before = samples collected before rain, after = samples collected after rain



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EFFECT OF PHENOLOGY AND RAINFALL ON ALLELOPATHY OF *XANTHIUM ITALICUM* MOR.

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The cocklebur species become more and more significant weed species in Hungary, where the row crops (such as: corn, sunflower, sugar beet, etc) are endangered by them very badly. Their fast spreading and danger are caused by many reasons: lasted sprouting, reduced sensitivity against many kinds of herbicides, vigorous competitional ability, allelopathy, fast initial growth and changes in climate. Allelopathy is used by many species of plants in the competition with one other, as well as by the cockleburs. But the efficiency of this is influenced by many factors. Species with allelopathical effect could have different attitudes under different conditions: they can produce materials with retarding and stimulating effects in different quantities or composition, and the actual conditions of existences, of acceptor plants and the mediator agent (i.e. soil) influence the final effect. In this experiments influence of phenology and rainfall were studied on allelopathy of cockleburs in May and July. In July a stronger inhibitor effect was observed on growth of cress before rain than in May, but it disappeared after rain. Inhibition of sugarbeet's germination was stronger in May than in July, and the difference became larger after rain.

STUDY ON THE NUTRIENT CONTENT OF WEEDS IN POTATO

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In the past years weed flora has changed significantly. Some species have spreaded while other ones have disappeared. In the course of permanent usage of herbicides weed population of cultivated fields has changed disadvantageously: tolerant and resistant weed breeds have appeared and spreaded (Reisinger 2001). Each crop has own special weed flora, which can be influenced by several factors, for example nutrients (Lehoczky 1995).

Lots of weeds can be found in potato crop. From the point of view of importance *Chenopodium album* L. is on the first place (Maykuss 1993). This is one of our most spreaded weed breeds, which can be found not only the fields, but in every crop culture (Ujvárosi 1973). Based on the data of IV. National Weed Survey *C. album* is on the 4th place (Tóth-Spilák 1998).

Echinochloa crus-galli (L.) P.B. and *Amaranthus* spp. have the same signification. The 900 species of the *Amaranthaceae* family are spreaded all over the world, except the cold zone (Ujvárosi 1973). *Amaranthus retroflexus* L. is on the 3th place while *Amaranthus chlorostachys* WILLD. has the 9th place on the weed dominance order in Hungary (Tóth-Spilák 1998). *E. crus-galli* is the dangerous weed of several crops. It is a cosmopolitan species (Lehoczky 2002). Based on the results of the IV. National Weed Survey *E. crus-galli* on the 2th place (Tóth-Spilák 1993). Similar to potato *Solanum nigrum* L. is belonging to the *Solanaceae* family so its weed control is different (Hoffmanné 2002).

Potato needs 4 kg N, 2 kg P_2O_5 and 8 kg K_2O for 1 t potato tubers (Horváth 1997). For 30 t tubers potato needs 150 kg N, 60 kg P_2O_5, 270 kg K_2O, 90 kg CaO and 30 kg MgO (Antal 1996).

At the early stage of development potato cannot compete with the weeds, thus proper control has a special importance. The quantity and forms of the damages caused by weeds can be a lot of kind. At the early stage of development weeds can use the water, light and space but the nutrients (Lehoczky 2002, Lehoczky and Reisinger 2002). Successful weed control has several factors for example the knowing of weed species and selection of effective herbicides (Béres 2000, Reisinger 2000). Competition between

weeds and crop is for the essential factors, for example water, light and nutrients. The significant nutrient usage of the weeds can cause disadvantageous effect on the crops (Lehoczky 1988, 1994, Kazinczi 1993, 1998).
On the effect of competition the quantity and quality of the crop increase that is caused by the lack of the water and nutrient used by weeds.
Our aim was the study of the nutrient concentration of the weeds in potato with the examination of the weight of the biomass and nutrient uptake of the plants.

Materials and methods

Our examination on competition was made in a field trial. The experiment was set up on the plots of Veszprém University, Georgikon Faculty of Agriculture, Regional Potato Research Centre. A randomised blocks design with four replicates and 27.6 m^2 plots was used. Twenty-eight potato tubers cv. „White Lady" were planted in a row. In autumn before planting and at the time of planting, fertilizers were applied. In autumn 200 kg $N \cdot ha^{-1}$ was applied. In spring 80 kg $N \cdot ha^{-1}$, 16 kg $P \cdot ha^{-1}$ and 24 kg $K \cdot ha^{-1}$ were applied at the time of planting. The quantities of the applied fertilizers and the time of application are the part of the crop technology of the Regional Potato Research Centre. On the untreated control plots there was not weed control from the planting of potato (23 April 2002) until harvest. On these plots we could examined the nutrient uptake of potato and weeds.
Potato and weed samples were collected on 18 June, after flowering of potato. Weeds were collected as species from 1 m^2 sampling area of the plots. Fresh and dry weight of the weeds and potato (shoots and tubers) were measured. N, P, K and Ca concentration of the samples was analyzed. Mathematical-statistical analyzes of the experimental date was made by SPSS software.

Results

Twelve weed species were found on the weedy control plots (Table 1).
From among these weeds *S. nigrum*, *A. chlorostachys*, *A. theophrasti*, *C. album* and *A. artemisiifolia* were the most important species. Weeds with T_4 life cycle form were dominated. There were two perennial weeds on the experimental plots: *C. arvensis* and *L. tuberosus.*

Table 1. Weed species occurring on the untreated (weedy) plots

Weed species	Code
Abutilon theophrasti MEDIC.	ABUTH
Amaranthus chlorostachys WILLD.	AMACH
Ambrosia artemisiifolia L.	AMBAR
Chenopodium album L.	CHEAL
Convolvulus arvensis L.	CONAR
Echinochloa crus-galli (L.) P.B.	ECHCG
*Lathyrus tuberosu*s L.	LATTU
Matricaria inodora L.	MATIN
Polygonum aviculare L.	POLAV
Polygonum lapathifolium L.	POLLA
Setaria glauca (L.) P.B.	SETGL
Solanum nigrum L.	SOLNI

Fresh weed shoot weight was higher than fresh potato shoot weight (Table 2,3). Comparing the fresh potato weight to the fresh weed weight we have found that the weight of the weeds is 30% of the potato weight. This fact shows that weeds were able to spread in high mass. Comparing the dry weed weight to the dry potato weight we have found that there is a difference between the results of fresh and dry weight. Dry matter of the weeds was more by 9% than the dry weight of potato (Table 2,3).

Table 2. Fresh and dry weight of potato on the untreated (weedy) plots ($g\ m^{-2}$)

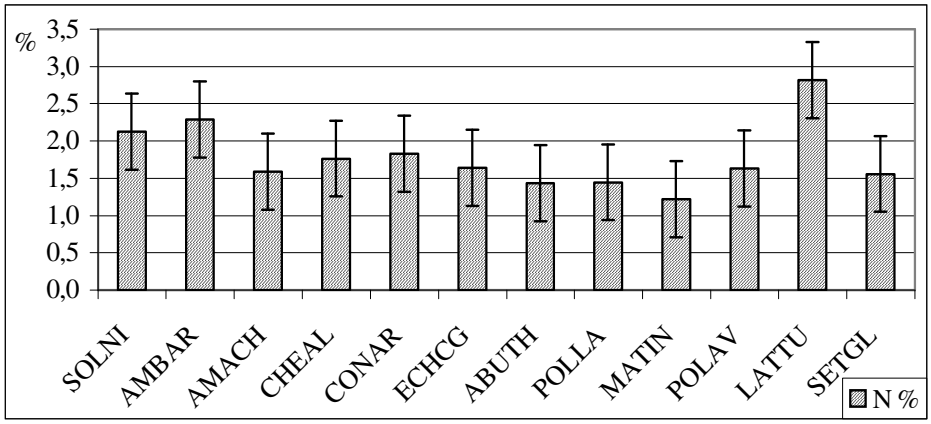
Potato	Fresh weight	Dry weight
Tuber	1650	170
Shoot	295	52
Total	1945	222

Table 3. Fresh and dry weight of the weed shoots (g m^{-2})

Weed species	Fresh weight	Dry weight	Water %
Solanum nigrum L.	283,8	50,9	82,1
Amaranthus chlorostachys WILLD.	158,8	38,7	75,6
Abutilon theophrasti MEDIC.	102,9	35,8	65,1
Chenopodium album L.	114,6	35,0	69,4
Ambrosia artemisiifolia L.	126,6	30,5	75,9
Polygonum lapathifolium L.	87,9	23,1	73,6
Convolvulus arvensis L.	40,0	9,7	75,6
Echinochloa crus-galli (L.) P.B.	29,8	6,3	78,6
Polygonum aviculare L.	18,4	5,8	68,1
Matricaria inodora L.	11,4	3,2	71,5
Lathyrus tuberosus L.	8,3	1,5	81,3
Setaria glauca (L.) P.B.	8,2	1,3	83,5
Total	991,3	242,5	-

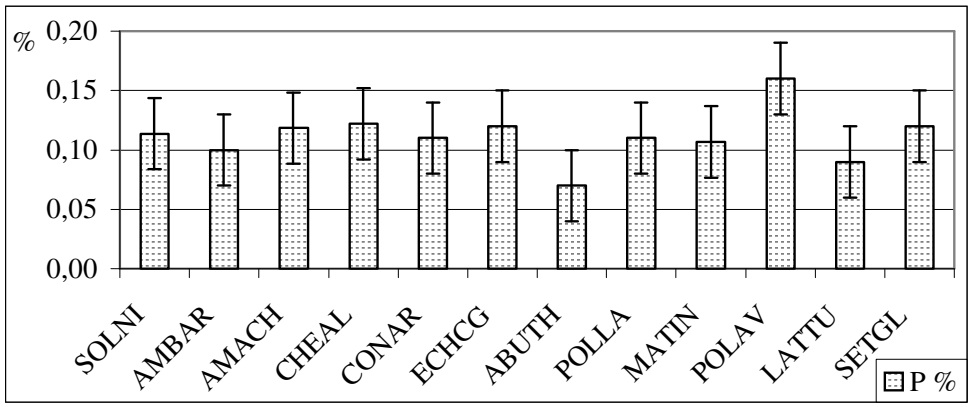
Water content of potato tubers and shoot altogether was 88.6% while water content of weeds was 75.6%. In the point of view of competition it is important to know the water content of the weeds. *S. glauca*, *S. nigrum* and *E. crus-galli* content a lot of water while *A. theophrasti*, *P. aviculare* and *C. album* had less water. Nutrient concentration and uptake by the plants were measured. N concentration of the weed shoots was 1.2-2.8% (Fig. 1).

Figure 1. Nitrogen concentration of the weed shoots, in the % of dry matter



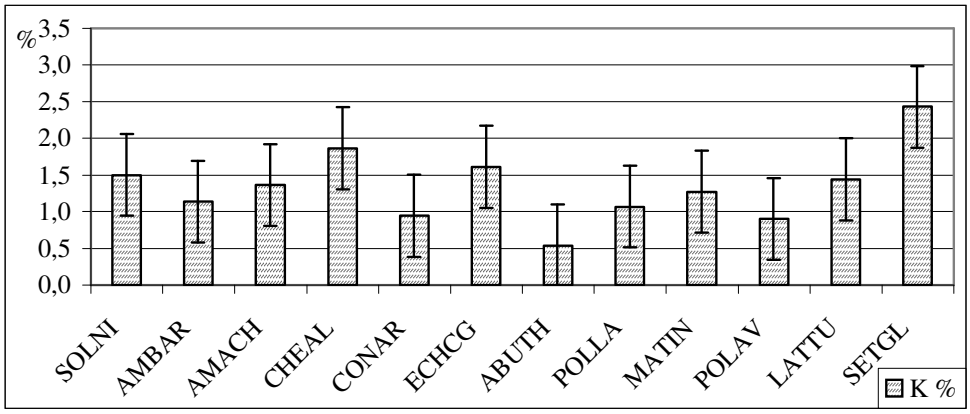
This nutrient could be found in the highest amount in the weeds. In the shoots of *L. tuberosus* N concentration was especially high, but *A. artemisiifolia*, *S. nigrum*, *C. arvensis* and *C. album* have a lot of N, too. In the half of the examined weed species N concentration was 1.5-2.0%. N content of potato shoots was 1.91% that is similar to the weeds. P concentration of the weeds was 0.06-0.16% (Fig. 2).

Figure 2. Phosphorus concentration of the weed shoots, in the % of dry matter



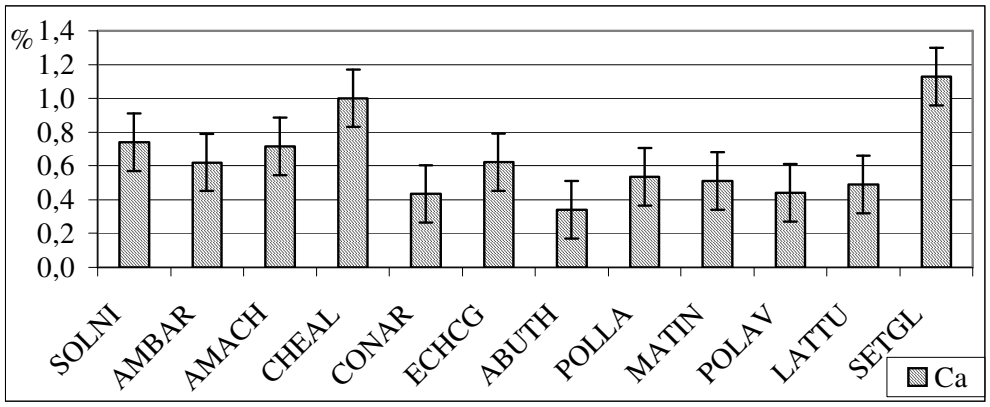
From among the measured nutrients P could be found in the lowest amount in the plants. P concentration of potato tubers and shoots was higher than in the weeds, 0.3%. Similar to the nitrogen, K concentration could be found in high concentration in the plants. The measured K concentration was 0.54-2.43% (Fig. 3).

Figure 3. Potassium concentration of the weed shoots, in the % of dry matter



From among the weeds in *S. glauca*, *C. album*, *E. crus-galli* and *S. nigrum* K concentration was very high. In potato shoots K concentration was 3.01% while tubers content 2.10% K. It is known that potato takes up K in the highest amount. Ca concentration of the weeds was between 0.34-1.13% (Fig. 4).

Figure 4. Calcium concentration of the weed shoots, in the % of dry matter



In the shoots of *S. glauca*, *C. album*, *S. nigrum* and *A. artemisiifolia* Ca concentration was high. Ca concentration in potato shoots was 3.03% while tubers have 0.97% Ca.
The amounts of nutrients taken up by plants until the examination were counted. Competition for N was very strong weeds have taken up more N by 30% than potato plants. From among weed species *S. nigrum* has taken up the most N, but *A. artemisiifolia* and *A. chlorostachys* have taken up a lot of N, too. These three weed species are „nitrophyl" species.
The amount of P taken up by plants was less than N. On the weedy plots potato has taken up the 57% of P, while the amount of P taken up by weeds was 43%.
Potato plants have taken up K in the highest amount from among the examined nutrients. The effect of competition can be seen in the K uptake of potato. Plants have taken up 7.4 g K altogether from 1 m^2. Potato has taken up 58% K and weeds have taken up 42% K (Table 4).

Table 4. The amounts of nutrient elements taken up by weeds and potato on the weedy plots (mg m^{-2})

Weed species	N	P	K	Ca
SOLNI	1172,8	54,1	711,7	360,1
ABUTH	700,0	31,2	345,8	121,6
AMACH	640,0	47,1	547,8	283,6
AMBAR	510,0	25,5	190,9	190,7
CHEAL	490,0	44,9	690,9	345,0
POLLA	330,0	24,0	240,7	121,5
CONAR	190,0	11,4	112,0	46,5
ECHCG	106,6	7,9	110,6	40,5
POLAV	100,0	9,6	49,8	28,6
LATTU	43,7	1,3	24,9	7,2
MATIN	40,0	3,2	41,5	14,3
SETGL	20,0	1,7	33,2	14,3
Total (weeds)	4309	262	3100	1573
Potato	3339	303	4278	2333

Based on literature data potato needs 3 kg Ca to 1 t tubers. From among the 12 weed species four species have taken up 70% of the Ca taken

up by all weeds. These species are as follows: *S. nigrum, C. album, A. chlorostachys* and *A. artemisiifolia*.

Conclusions

Based on our experimental data we have found that on the untreated weedy plots the spreading of the weeds was very high. Weed fresh weight was 50% of the potato fresh weight.

Weeds have taken up 4.35 g N, 0.26 g P, 3.10 g K and 1.57 g Ca altogether. These nutrient quantities correspond to 43.5 kg N ha^{-1}, 2.6 kg P ha^{-1}, 31 kg K ha^{-1} and 15.7 kg Ca ha^{-1}.

Acknowledgement

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NEW POSSIBILITIES IN THE SUNFLOWER POSTEMERGENT HERBICIDE APPLICATION

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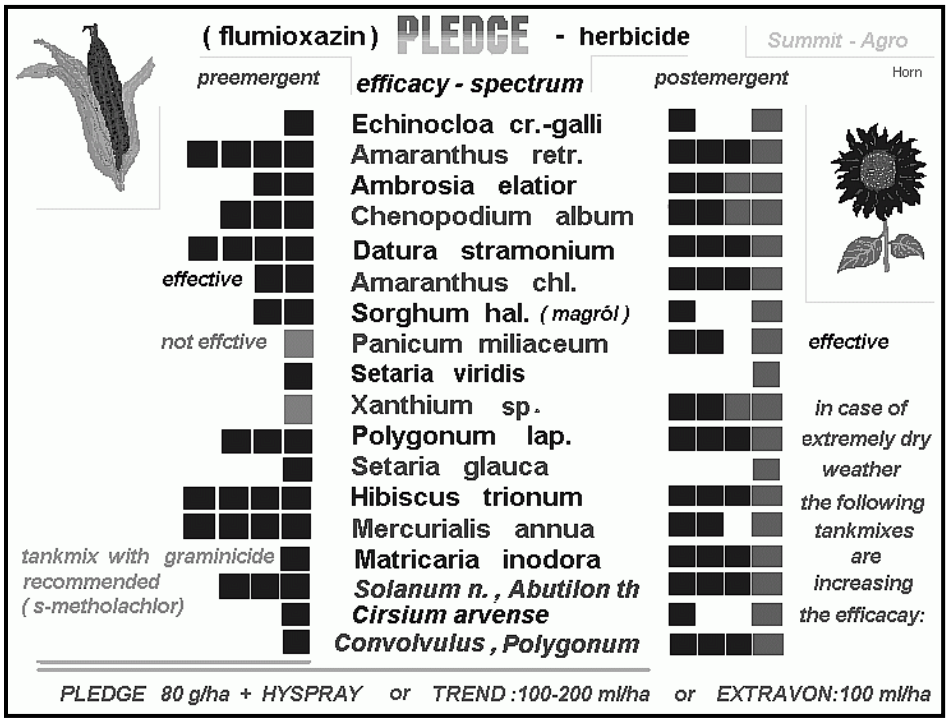
Flumioxazine (PLEDGE-SUMISOYA) is a well known herbicide in the broad leave-control of soybean and maize (corn) all over the world.
Main properties of flumioxazine (PLEDGE) are follows:

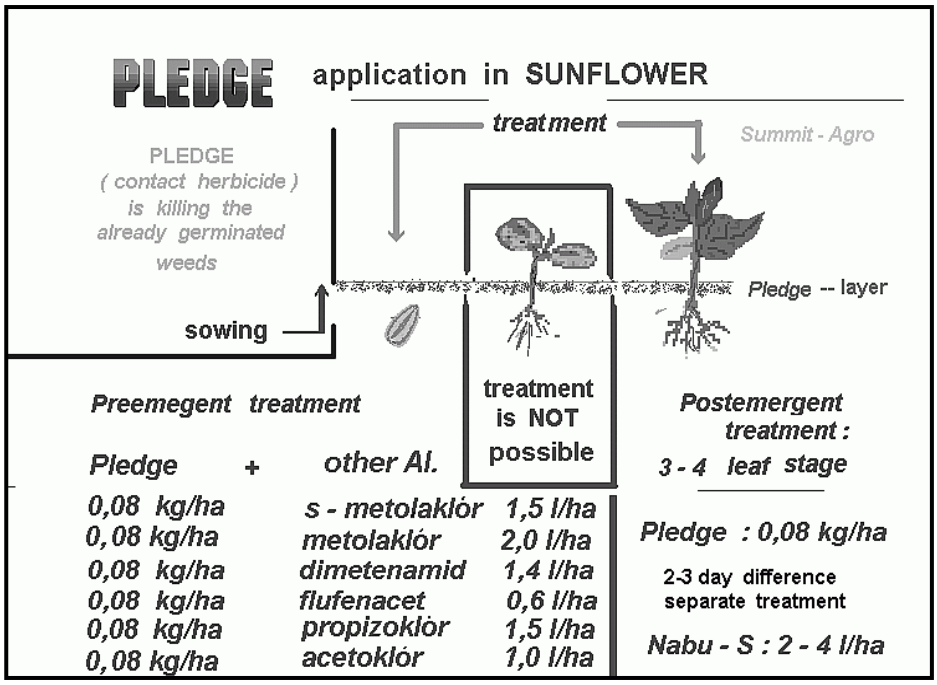
- efficacy is independent from the temperature.
- in case of pre- or postemergent application 5-10 mm rainfall after the treatment increases the effect of the herbicide.
- no phytotoxicity is on the following crop. The half-life time (degradation) is 30 days under average conditions. Organic matter content of the soil has limited influence on the herbicide efficacy.
- The product has very low water-dilution capability.

Figure 1. Main weeds in sunflower and effect of flumioxazine on them treated preemergence or postemergence



As a results of a 6 years development work of the trading company Summit-Agro Hungaria, the product was registered in Hungary in sunflower as a preemergent and postemergent herbicide as well.
In Figure1 the main weeds in sunflower (ranking of the weeds) and the effect of flumioxazine as preemergent and postemergent treatment are summarized.
In Figure 2 are summarized the tankmixture possibilities and dosages in sunflower.

Figure 2. Tankmixture possibilities and dosages in sunflower



AUTUMN APPLICATION OF METSULFURON-METHYL AGAINST *APERA SPICA-VENTI* (L.) P.B.

Elemér Tóth[1] – István Molnár[1] – István Somlyay[1] – Sándor Bálint[1] – Margit Nagy[2] – Lajos Szőke[2]

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Application of autumn weed control against T_1 and T_2 life cycle type weed species is a tradition on some part of Western Europe mainly against *Alopecurus myosuroides* and *Apera spica-venti*. In Hungary there is a very limited infection of *Alopecurus myosuroides*, but *Apera spica–venti* is a serious problem on acidic soils. Its spreading areas are Szabolcs-Szatmár-Bereg, Somogy, Zala, Vas, Győr-Moson-Sopron and Veszpém counties. In 2003 metsulfuron-methyl (Ally® 20 DF), a new herbicide was registered in winter wheat, winter barley and tricitale in Hungary. In Western Europe a lot of farmers are using this herbicide to control black grass (*Alopecurus myosuroides*), but there is not too much data how it can control loose silky bent (*Apera spica-venti)*. It was the reason, why a trial was carried out using of Ally® 20 DF on *Apera* infested area in Hungary.

Literature

There cannot be found a publication in the international publication lists which is written in this topic (connection of metsulfuron and *Apera spica-venti*), but there are some publications speaking about control of this weed species by other active ingredients.
Prometryne + simazine was examined at the eastern part of Germany (BUHR et al., 1977). Primary dormancy of seeds of *Apera spica-venti* and *Alopecurus myosuroides* was tested by WALLGREN and AVHOLM (1978) and demonstrated that both weed species show a typical winter annual habit with a germination peak in the autumn and little or none during the following spring after the seeds had been „stored" outdoors in the soil.
In Poland efficacy of some treatments (MCPA+ dicamba, 2,4-D+ dicamba, amidosulfuron, isoproturon, chlorsulfuron, fenoxaprop-P+ isoproturon) was compared by DOMARADZKI and ROLA (1997). The best efficacy was given by chlorsulfuron. The emerging of loose silky bent on sandy soils was proven by PETERSEN in 1998 in Denmark, where the early drilling resulted in heavy loose silky bent emergence. Even though loose silky bent

is controlled, the greatest yield are achieved after late drilling. Field studies were conducted in Poland during 1998 to study the effectiveness of a combined application of herbicide (Chisel 75 DF)+ growth regulator+ adjuvant mixture. The control of *Apera spica-venti* increased by 4-9 %. There was proven by CIMERMAN and BABNIK (1999) that isoproturon active ingredient acts residually for 2-3 months, residual activity of amidosulfuron is shown for 1-3 weeks against *Apera spica-venti*. Also ROLA et al. (1999) studied the efficacy of some sulfonylurea herbicides (chlorsulfuron + thifensulfuron-methyl, rimsulfuron and triflusulfuron) in tank mixture with adjuvants in winter wheat, corn and sugar beet. KLEM and VANOVA (2000) were examined the effect of sulfosulfuron on *Apera spica-venti* and *Elytrigia (Elymus, Agropyron) repens*.

Materials and Methods

The efficacy of metsulfuron-methyl against *Apera spica-venti* was tested by Plant and Soil Protection Service of Szabolcs-Szatmár-Bereg county in Nagyhalász, Hungary in 2002/2003 season. Date of treatments were the following: autumn postemergence treatment: November 18, 2002, spring postemergence treatment April 15, 2003. The trial was treated by a "Szolnok" type sprayer machine, water volume was 300 litre/ha, pressure 2 bar. Plot size: 20 m^2 , number of replications: 4.
In Nagyhalász there is a brown forest type of soil, which has 1.81 % organic matter, this soil is acidic: pH(KCl) = 5,13.
The crop before winter wheat was in the 2001/2002 season was also winter wheat, the stubble-field peeling was done at July 26, 2003, then the soil preparation before sowing was carried out at October 8, 2003. Altogether – because the owner of field had some machinery capacity problem- the quality of soil preparation could be only sufficient.
Sowing of winter wheat was made at October 21, 2003, variety was GK Petur. In October the trial territory got 78 mm precipitation, after it *Apera spica-venti* started to emerge. So this fact resulted that by the time of the autumn postemegence treatment the phenology of *Apera spica-venti* was 1-3 (4) leaves stage, *Tripleurospermum inodorum (Matricaria inodora)* cotyledon- 2 leaves stage. Winter wheat was by the time of autumn postemergence treatment 1-3 leaves stage, by the time of spring postemergence treatments in tillering. By spring postemergence treatments *Apera spica-venti* individuals were foundable from 1 leaves stage upto tillering, *Tripleurospermum inodorum (Matricaria inodora)* was in 2-4 (6) leaves stage.

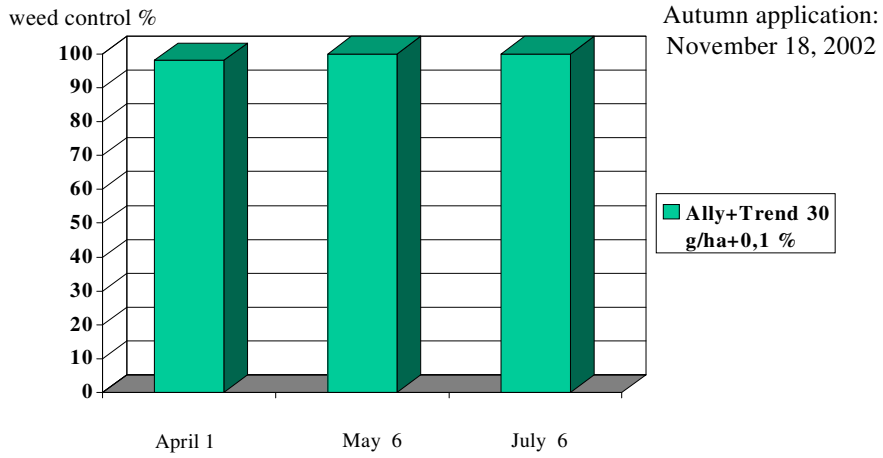
In four weeks after the autumn postemergence treatemnts the territory got 33 mm precipitation, in two weeks after the spring postemergence treatments 10 mm, but in four weeks also 33 mm precipitation.
Assessment were done three times: the first assessment at April 1, 2003; second assessment at May 6, 2003; third assessment at July 6, 2003 (just before harvesting).

Results

Effect of Ally® 30 g/ha + Trend™ 0,1 % autumn postemergence treatment gave against in autumn emerged *Apera spica-venti* plants excellent weed control and this treatment could prohibit the spring emerge of this weed species.
Autumn application of Ally® 30 g/ha + Trend™ 0,1 % treatment proved its excellent efficacy against loose silky bent (Figure 1), and ensured 100 % efficacy against *Tripleurospermum inodorum* in case of all of the three assessments.

Figure 1.

Winter wheat weed control trial against *Apera spica-venti*.
Crop and Soil Protection Service of Szabolcs-Szatmár-Bereg county, Nagyhalász, Hungary 2003.

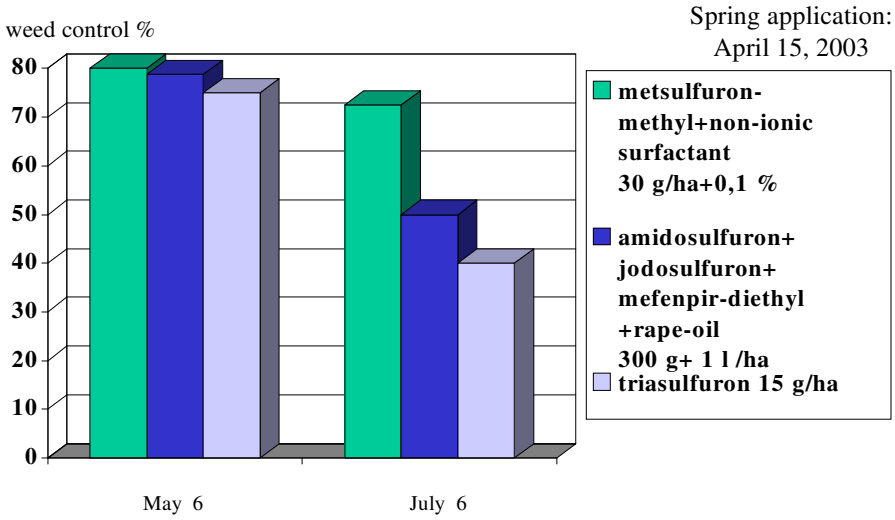


In the springtime a new flush of *Apera spica-venti* started to emerge, and the spring was very windy, that were the reasons, why the spring postemergence treatments were made at April 15. That time some individuals of loose silky bent emerged in autumn were already after tillering.

The effect of spring application of Ally® 30 g/ha + Trend™ 0,1 % treatment controlled well the less developed *Apera spica-venti* plants among the autumn emerged individuals three weeks after the treatment, plants of loose silky bent, which were more developed than 4-5 leaves stage by the spring treatment, were damaged, but some of them were regenerated in July.
There could be establish that spring application of metsulfuron-methyl + non-ionic surfactant in this case could not give enough (more than 90 %) efficacy against the heterogen *Apera spica-venti* population, but showed better result than the standard treatments (amidosulfuron + jodosulfuron + mefenpir-diethyl + rape-oil 300 g + 1 l/ha and triasulfuron 15 g/ha). Metsulfuron-methyl could give longer effect than the standard treatments (Figure 2).

Figure 2.

Winter wheat weed control trial against *Apera spica-venti*.
Crop and Soil Protection Service of Szabolcs-Szatmár-Bereg county, Nagyhalász, Hungary 2003.



Discussions

Autumn use of Ally® 30 g/ha + Trend™ 0,1 % treatment showed an excellent weed control against *Apera-spica-venti* and it could keep that efficacy up to harvesting (Figure 3). The product is registered in Hungary from 3 leaves stage up to end of tillering of winter wheat, winter barley and triticale crops.

Figure 3. Nagyhalász, Eastern Hungary, 2003.

left: untreated right: Ally® 30 g/ha + Trend™ 0,1 % (autumn treatment)

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Summary

AUTUMN APPLICATION OF METSULFURON-METHYL AGAINST *APERA SPICA-VENTI* (L.) P.B.

E. Tóth[1], I. Molnár[1], I. Somlyay[1], S. Bálint[1], M. Nagy[2] and L. Szőke[2]

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[2] Crop and Soil Protection Service of Szabolcs-Szatmár-Bereg county, Nyíregyháza, Hungary

Autumn application of metsulfuron-methyl 30 g f.p./ha + non-ionic surfactant 0.1 % was examined against *Apera spica-venti* in Hungary. The treatment showed an excellent weed control against loose silky bent and it could keep that efficacy up to harvesting. The product is registered in Hungary from 3 leaves stage up to end of tillering of winter wheat, winter barley and triticale crops.

INDUCTION OF MAIZE GLUTATHIONE S-TRANSFERASES BY HALOACETAL, HALOKETAL AND HALOAMIDE SAFENERS

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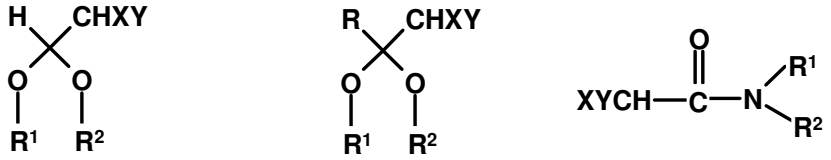
Acetochlor (**Ac,** 2-chloro-N-ethoxymethyl-N-(2-ethyl-6-methylphenyl) acetamide) is a chloroacetanilide herbicide applied preemergence to control many annual grass and certain broadleaf weeds in maize (*Zea mays* L.). Maize exhibits marginal tolerance to the acetochlor. Therefore, the use of safeners is neccessary to overcome the phytotoxic symptoms such as shoot and root growth reduction as well as distorted leaves and coleoptiles as consequences of the acetochlor treatment.

Safeners are chemical agents that increase the tolerance of crop plants to herbicides without affecting the weed control efficacy. They appear to induce a set of genes that encode enzymes and the biosynthesis of cofactors involved in the herbicide detoxicaton (Gatz, 1997). Glutathione S-transferase isoenzymes (GSTs) and endogenous glutathione (GSH) play a vital role in chloroacetanilide herbicide detoxication by GSH conjugation. Safeners of various chemical classes were found to induce the activity of GSTs and the level of GSH in the protected plants (Davies and Caseley, 1999). The dichloromethyl ketal MG-191 (2-dichloromethyl-2-methyl-1,3-dioxolane) is highly active used in the safening of maize against thiocarbamate and to a lesser extent chloroacetamide herbicides (Jablonkai, *et al*., 2001). MG-191 similarly to the other safeners antagonizes the growth inhibition of the acetochlor by eliminating the shoot-twisting and leaf-rolling injuries of maize seedlings but appears to be less effective in the root system even (Jablonkai, 1991).

In order to further clarify the mode of action of MG-191 and analogous molecules the significance of GST and GSH enhancement in safening maize against the herbicide acetochlor, the relationship of structure to safening efficacy, GSH and GST inducibility was examined using halogenated acetals (**1a-l**), ketals (**2a-k**) and acetamides (**3a-d**) with emphasis on the roots (Figure 1).

Figure 1. Examined halogenated acetals (details in the text)



1a-l **2a-k** **3a-d**

Materials and Methods

Chemicals

Open-chain dichloromethyl acetals and ketals were synthesized from dichloroacetaldehyde, 1,1-dichloroacetone, and 1,1-dichloroacetophenone (Dutka, 1991). Cyclic acetals and ketals were prepared from diethyl acetal and ketal of dichloroacetaldehyde and 1,1-dichloroacetone by transacetalisation. Acetamides were synthesised by haloacetylation of amines using standard Schotten-Baumann conditions. Crude reaction products were purified by either distillation or silica gel column chromatography. Acetochlor was purified by column chromatography from the commercial product. [Carbonyl-^{14}C]acetochlor (sp. act. 37 MBq /mmol) was a sample prepared previously (Jablonkai and Hatzios, 1991).

Safener activity of experimental molecules

Seeds of maize (Gazda Martonvasar, Hungary) were soaked in water and planted in plastic cups (6 cm diameter, 9 cm deep, 3 seeds/cup) containing air-dried foundry sand (250 g, OH-4 type). Treatment solutions (50 ml) containing safener (50 μM) and/or acetochlor (50 μM) were applied to each cup. Seeds were placed 2 cm deep. The plants were grown in a growth room (temperature: 23 $\pm$ 1 ^{o}C; relative humidity: 60 $\pm$ 5 %; light intensity: 10 klux; light period: 16 h per day). The plants were watered three times a week to bring the weight of cups to 300 g. Plants were harvested two weeks after the treatment shoot and root lenghts measured. The experiment was carried out twice with four replicates.

Plant materials and enzyme isolation

For GST activity analyses seeds (25) of maize were placed in Petri dishes (18.5 cm in diameter) on two layers of filter-paper wetted by aqueous

solution (20 ml, 50 μM) of chemicals studied. The dishes were placed in a germination thermostat. The seedlings were grown in the dark for 5 days at 27 ^{o}C. Five-day-old seedlings were thoroughly washed with tap water and separated roots were homogenized in a mortar and pestle using quartz sand then extracted with 5 volumes of cold Tris-HCl buffer (100 mM, pH 7.5) containing 2 mM EDTA, 1 mM dithiothreitol and 5 % (w/v) polivinyl polypirrolidone. The homogenates were filtered through two layers of Miracloth and the filtrates were centrifuged at 10,000 x g for 20 min at 4 ^{o}C. The supernatants were brought to 80% $(NH_4)_2SO_4$ to 80 % saturation and centrifuged at 10,000 x g for 20 min at 4 ^{o}C. Aliqots of the protein precipitates were resuspended in potassium phosphate buffer (20 mM pH 6.5) and desalted by gel filtration (Sephadex G25, medium) before use for enzymatic studies.

For determination of GSH contents root tissues of etiolated seedlings were grown as described earlier. Tissues were frozen and homogenized in liquid nitrogen and extracted with 4 volumes of 70% ethanol The homogenates were centrifuged at 10,000 x g for 20 min at 4 ^{o}C and the supernatants were collected.

Analysis of GSTs and GSH

Glutathione *S*-transferase activities of desalted enzymes were determined with CDNB (1-chloro-2,4-dinitrobenzene) and [carbonyl-^{14}C]acetochlor (Ac) substrates. GST(CDNB) activities were determined spectrophotometrically (340 nm) and expressed as nmol product formed per second (nkat) per mg protein (Dixon *et al.*, 1998a). GST(Ac) activities of the samples were determined by liquid scintillation counting radioassaying the conjugate formed in the reaction of [carbonyl-^{14}C]acetochlor (0.75 mM) and GSH (10.0 mM) mediated by the desalted enzymes at 37 ^{o}C in 30 min. The GST(Ac) activity was expressed as pmol conjugate per second (pkat) per mg protein. Protein contents of the extracts were determined spectrophotometrically using a Coomassie Brillant Blue reagent with bovine serum albumin as reference protein.

Non-protein thiol (GSH) content of alcoholic supernatant was measured spectrophotomet-rically (412 nm) using DTNB reagent (Jablonkai and Hatzios, 1991).

Results and Discussion

Safening experiments were carried out in sand at a relatively high pre-emergence acetochlor rate (2.4 kg/ha) and at high moisture content. Under these conditions the herbicide is extremely phytotoxic and no complete

protection can be achieved. The protection of shoots by bromoacetaldehyde diethyl acetal (**1b**) was moderate while the monochloroacetal **1a** and the dichloroacetals (**1c-l**) exhibited poor or no safening activity (Table 1). Among dialkyl ketals having increasing alkyl chain length (**2a-d**) the highest safening activity was observed for the diethyl (**2a**) and dipropyl (**2b**) derivative. In general, cyclic ketals (**2e-k**) were effective safeners.

Derivatives having 1,3-dioxolane (**2f**), dioxane (**2g**) and dioxepane (**2i**) ring in their structure were the most active molecules. Interestingly, dioxacycloalkanes with 8- and 9-membered ring were still active in safening in the shoot zone. The safening activity of ketals also exceeded that of acetals against the thiocarbamate EPTC (Dutka, 1991). Among amides the marketed safener dichloroacetyl-diallylamide (dichlormid, **3a**) was highly protective and decreasing the number of halogens and allyl groups yielded less active molecules. The root growth inhibition of the acetochlor was less antagonised by the ketals **2a-k** and the amides **3a-d** as compared to safening the shoots of maize seedlings (Table 1). However the protective action of the acetals **1c-l** in the root zone was superior to that in shoot zone and exceeded the root safening activity of the ketals and the amides. The higher activity of the acetals in the roots can be explained by their increased root uptake and reduced mobility towards shoots. In an early study the ketal type MG-191 (**2f**) was found extremely mobile after root application and its enhanced mobility to the root-absorbed acetochlor was considered as an important factor in its safening efficacy (Jablonkai, 1991).

The GSH content of the root tissues of safener-treated plants was only slightly affected by treatment with either acetals or ketals as compared to that of untreated control (Figure 1). The only exception was the moderately safening **2e** cyclic ketal that triggered out a 2.4-fold increase. Cyclic acetals were inhibitory of GSH biosynthesis in roots. Among amides a high degree of induction was observed for the less effective safener **3b.** It seems that no correlation exists between the elevation of GSH content in roots and the shoot safening efficacy of these molecules. On the other hands the elevation of GSH content has been observed for a number of safeners (Davies and Casely, 1999).

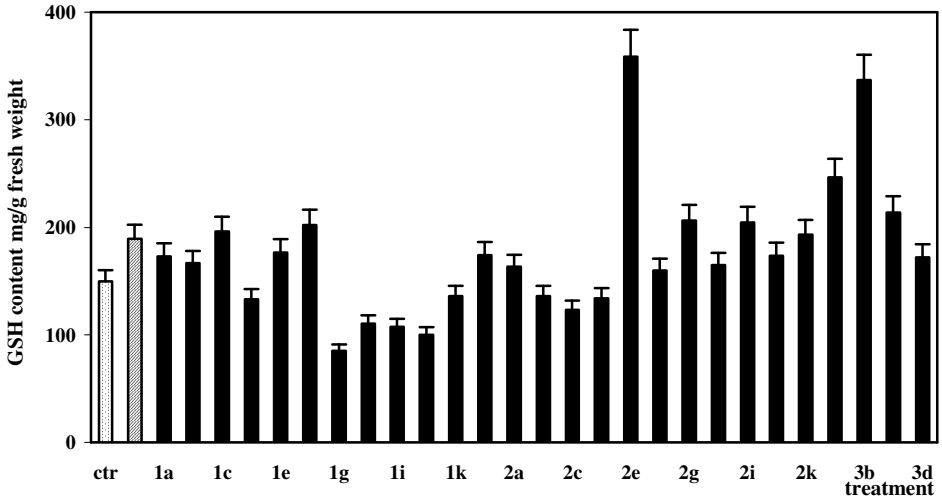
Table 1. Structure and safening activity of acetals, ketals and amides towards the acetochlor in maize

Code	R	X	Y	R^1	R^2	Protection of shoots[a] (%)	Protection of roots[b] (%)
1a	-	H	Cl	Et	Et	24	-46
1b	-	H	Br	Et	Et	60	-12
1c	-	Cl	Cl	Et	Et	8	33
1d	-	Cl	Cl	Pr	Pr	0	55
1e	-	Cl	Cl	Bu	Bu	-6	12
1f	-	Cl	Cl	i-Bu	i-Bu	-2	18
1g	-	Cl	Cl	$-(CH_2)_2-$		18	48
1h	-	Cl	Cl	$-(CH_2)_3-$		14	3
1i	-	Cl	Cl	$-CH_2C(CH_3)_2CH_2-$		-3	33
1j	-	Cl	Cl	$-(CH_2)_4-$		11	39
1k	-	Cl	Cl	$-(CH_2)_5-$		0	9
1l	-	Cl	Cl	$-(CH_2)_6-$		3	42
2a	Me	Cl	Cl	Et	Et	62	11
2b	Me	Cl	Cl	Pr	Pr	63	14
2c	Me	Cl	Cl	Bu	Bu	38	20
2d	Me	Cl	Cl	i-Bu	i-Bu	14	8
2e	Ph	Cl	Cl	$-(CH_2)_2-$		41	45
2f	Me	Cl	Cl	$-(CH_2)_2-$		64	9
2g	Me	Cl	Cl	$-(CH_2)_3-$		68	11
2h	Me	Cl	Cl	$CH_2C(CH_3)_2CH_2-$		66	20
2i	Me	Cl	Cl	$-(CH_2)_4-$		70	28
2j	Me	Cl	Cl	$-(CH_2)_5-$		50	16
2k	Me	Cl	Cl	$-(CH_2)_6-$		60	45
3a	-	Cl	Cl	allyl	allyl	81	58
3b	-	H	Cl	H	allyl	48	23
3c	-	H	Cl	allyl	allyl	2	-92
3d	-	H	Br	allyl	allyl	22	12

[a] based on shoot length; protection (%) = 100 x [(herbicide + safener)] / [control - herbicide]; shoot lenghts 14 DAT: control, 27.9±5.3 cm, acetochlor, 3.1±0.3 cm

[b] based on root length; root lenghts 14 DAT: control, 9.5±1.2 cm, acetochlor, 6.2±1.2 cm

Figure 1. GSH content in the roots of maize seedlings pretreated with the acetochlor and the experimental safeners



GST(CDNB) activity of the root tissues was hardly affected by pretreatment with safeners (Figure 2). The acetals **1a-c** were inhibitory while treatment with **1k** and **1l** cyclic acetals showing no safening activity to the shoots and some activity to the roots resulted in 2- and 4-fold increase in this isoenzyme activity. In general, no relationship can be shown between their effects on GST(CDNB) activity and shoot or root safening efficacy. GST(CDNB) activity associated with the safener inducible *Zm*GSTF1-2 isozyme was increased by the dichlormid in roots and shoots of maize (Dixon et al., 1997) and only marginally effected by the MG-191 (Jablonkai et al., 2001).

GST(Ac) activity in the roots was enhanced by both protective and less effective structures as compared to that of untreated control (Fig. 3). The treatment with the herbicide acetochlor in itself doubled this isoenzyme activity and exceeded the effects of the acetals. Among ketals the safener MG-191 (**2f**) induced the highest increase in the enzyme activity while no elevation was shown for the molecule **2i** which posseses with the same safening activity in the shoot zone. For the amides the degree of induction on this isozyme activity appears to be parallel with their shoot safening potential. A correlation between GST(Ac) induction and safening activity of amides exists only in the shoot tissues (data not shown). These findings indicate that the mode of action of the acetals and ketals is likely differs from that of the amides.

Figure 2. GST(CDNB) activity in the roots of maize pretreated with the acetochlor and the experimental safeners

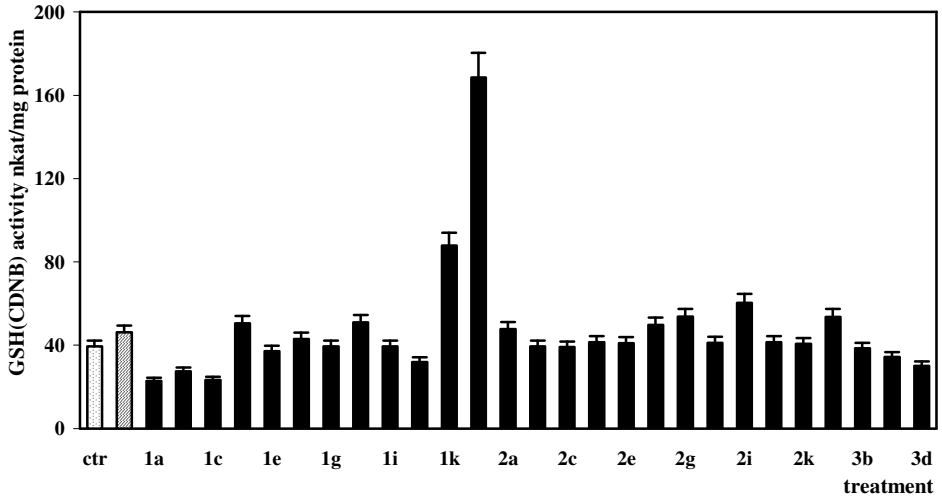
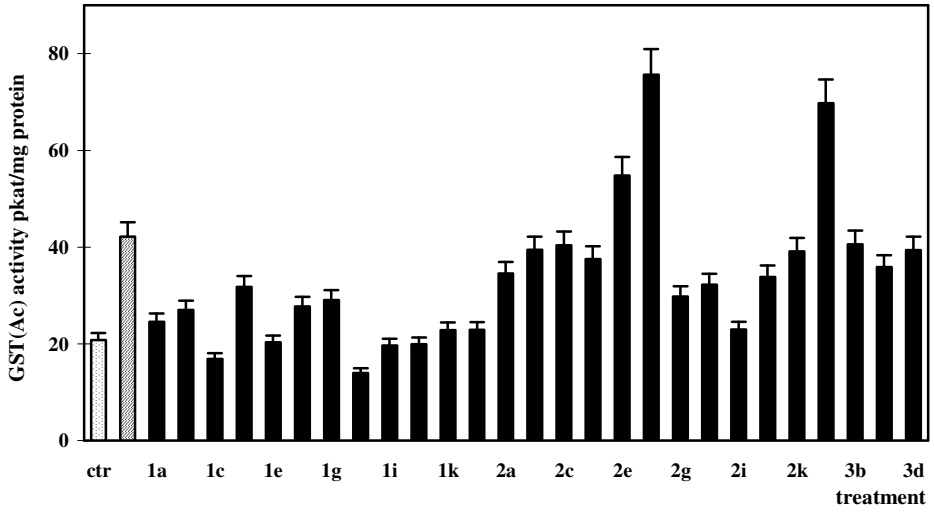


Figure 3. GST(Ac) activity in the roots of maize pretreated with the acetochlor and the experimental safeners



Pretreatment of maize seedlings with acetochlor resulted in a very high degree of induction of the enzyme activity indicating that the induction of GST isoforms by both chloroacetanilides and their safeners is based on a similar mechanism.

The exact mechanism of the safener-mediated enhancement of GST activity is not completely understood. GSTs are induced by a diverse range of chemicals and accompanied by the production of active oxygen species. Thus the connection between safener-mediated protection of crops and oxidative stress tolerance has been suggested (Theodoulou *et al.*, 2003). Many GSTs are effective not only in conjugating electrophilic substrates but also function as glutathione peroxidases. Safeners may induce GST expression by mimicking oxidative insult (Dixon *et al.* 1998b). Our results indicate that safener structure plays a decisive role in specific expression of GSTs mediating the detoxication of chloroacetamide herbicides and seems to be tissue specific. Since no strong correlation between the degree of induction of levels of GSH and GST isoforms and the safener activity was found the mode of action of safeners is a more complex process than simply promoting the metabolism of herbicides.

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Summary

INDUCTION OF MAIZE GLUTATHIONE S-TRANSFERASES BY HALOACETAL, HALOKETAL AND HALOAMIDE SAFENERS

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The herbicide safener MG-191 and its acetal and ketal analogues as well as mono-and dichloroacetamides were tested for their ability to alleviate toxicity of acetochlor to maize. The differential enhancement of the GSH content and the expression of GST isoforms was studied in roots of maize. Our results demonstrate that the safener structure affects the specific expression of GSTs mediating the detoxication of acetochlor. No correlation was found between the degree of induction of GSH and GSTs and the safening activity.

LECTURES OF ENTOMOLOGICAL & ECOLOGICAL SESSION

THE LACEWING FAUNA (NEUROPTERA: CHRYSOPIDAE) OF DEBRECEN

(Summary)

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Studies were conducted in the botanical garden and the surrounding experimental area of the Faculty of Agricultural Sciences in Debrecen to assess structure parameters of the occurring lacewing assemblage(s). Localities were sampled by sweeping net from early April until early October in 1996-2003.

In the sample area at least 12 chrysopid species have been captured among which 7 were found relatively constantly. Sibling species of the *Chrysoperla carnea* complex (*Chrysoperla affinis* (*kolthoffi*), *Chrysoperla carnea* s. str., *Chrysoperla lucasina*) were the most common species, comprising 75-96 % of the total, followed by individuals of *Dichochrysa prasina, Chrysopa pallens, Chrysopa formosa, Chrysopa perla, Chrysopa viridana* . *Chrysopa phyllochroma*, *Chrysopa nigricostata* and *Chrysotropia ciliata* specimens occurred only rarely and singly. The assemblages were characterized by low diversity and high similarity values. The original and processed data stress the scarcity of the lowland lacewing fauna.

SPILOSTETHUS [= LYGAEUS] EQUESTRIS L., (HETEROPTERA: LYGAEIDAE), A PEST OF SUNFLOWER

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According to Günther (1975), the *Spilostethus equestris* L. is indigenous from southern England to Siberia, and from central Sweden to the Mediterranean areas. It is less common in the north of Central Europe; it prefers areas with warmer climate. It likes staying both on the ground or in the flowers of various plants. It especially likes the tame poison or swallowwort (*Vincetoxicum officinale* MNCH., syn.: *Cynanchum vincetoxicum* (L.) PERS.).

Data on the life history of the *S. equestris* in Hungary is extremely scarce, despite the fact that the species is widespread in our country. Information on its nourishing plants and habits can only be found in the publications of Horváth (1984; 1986; 1987b; 1989; 1999) and Bujáki and Horváth (1992).

Horváth (1984) inferred the nourishing plant specialization of the species from the total nitrogen contents of various plant seeds, such as those of the *Asclepias syriaca* L., and the sunflower. Amino-acid composition in the seeds of the *A. syriaca* is the most similar to that of the sunflower, soy or peanut. It may be assumed that its similarity to the sunflower seed is the cause of the fact that the *S. equestris* willingly sucks the ripening achenes of the sunflower, too (Horváth, 1984).

Material and Methods

Our study was carried out in the districts of Bácsalmás („free" of *A. syriaca*) and Katymár (heavily infected with the *A. syriaca*) between 25 and 31 August 2001. The two districts were significantly different in respect of infection with *A. syriaca*.

While the *A. syriaca* only occurred sporadically in the forest stripes and in the edges of the industrial (hybrid propagating) sunflower fields in the Bácsalmás district, in the Katymár district the occurrence of the weed was 20-25 specimen/m^2.

The damage caused by *S. equestris* was investigated in the selected land strips as follows: ten randomly selected, adjacent sunflower discs were

examined at every 10 meters (10 locations in total) from the edge of the land strip towards the centre.
By carrying out these investigations we tried to assess the extent and frequency of the damage caused by the *S. equestris*; partly as a function of the number of the achenes damaged in a disc, and partly of the occurrence of the *A. syriaca*. We also wanted to understand the cause of the „greening of the seed inside the achene", which phenomenon adversely affects the export market position of the striped, alimentary purpose species (hybrids). Our investigations aimed at finding satisfactory answers both in respect of the market „condition" of the export consignments of the striped, alimentary purpose seeds, and the germination value (germ %) of hybrid sunflower propagation.

Results and Discussion

Both Table 1 and Table 2 show clearly the difference in the damage caused by the *S. equestris* that difference originates from the extent of infection with the *A. syriaca* (the main nourishing plant of the *S. equestris*). The occurrence of this insect significantly increased in the areas that were infected with the *A. syriaca* . Overwintering of the adults was relatively unhindered due to the mild winters in recent years.
The data in the tables make it unambiguous that the severe damage caused by plant bugs is prominent primarily in the 40 m wide strips of the land strips (Table 1 and 2). The settlement of, and characteristic damage caused by the bugs can best be observed in these stripes. This applies to the damage caharacteristics of both the Katymár district (which is more infected with the *A. syriaca*) and the less infected areas (Bácsalmás district). At the same time, it has also been proved, that now the damage caused by the *S. equestris* (which species was earlier referred to as a typical „pest of the edges" of land strips) (Horváth, 1989; 1991) is no longer restricted to the edges of the land strips. Although to a decreasing extent, but it is present at a distance of 100 metres from the edge of the land strip, that is worth paying attention. It is especially true, as the *S. equestris* may be a vector of the cucumber mosaic virus (*Cucumber mosaic Cucumber virus*) or other viruses, the presence of which can be demonstrated on the *A. syriaca* (Horváth, 1980; 1981). This is also supported by the observations of recent years that refer to the spreading of the damage caused by pathotype B of the cucumber mosaic virus (CMV), and the *sunflower chloratic virus* (SuCMoV) (Salamon, 2002, Lenardon et al., 2001). The severe damage caused by the bug locally (in spots within a strip of land) usually occurs after the fall of tubular flowers (about the middle or end of August).

Table 1. Occurrence of achenes damaged by *Spilostethus equestris L.* (number of specimen) Katymár, 2001

Number	Number of damaged achenes per location (10 discs from each location)										Total, pcs	Mean of 10 discs
	1	2	3	4	5	6	7	8	9	10		
	(10m)	(20m)	(30m)	(40m)	(50m)	(60m)	(70m)	(80m)	(90m)	(100m)		
1	47	50	36	20	20	16	16	15	19	8	247	24.7
2	59	63	40	60	10	20	20	10	21	16	319	31.9
3	65	50	38	30	15	25	12	16	20	20	291	29.1
4	70	48	45	40	16	16	13	10	14	5	278	27.8
5	96	44	47	48	17	29	10	14	20	26	351	35.1
6	20	60	62	35	32	30	15	22	26	10	312	31.2
7	34	35	34	46	38	26	24	20	20	11	288	28.8
8	75	70	46	45	30	30	26	18	13	16	369	36.9
9	60	46	40	60	26	31	32	26	16	18	355	35.5
10	70	49	39	32	24	48	40	30	20	11	361	36.1
Total:	**596**	**515**	**427**	**416**	**228**	**269**	**208**	**181**	**189**	**142**	3171	31.71

Table 2. Occurrence of achenes damaged by *Spilostethus equestris L.* (number of specimen) Bácsalmás, 2001

Number	Number of damaged achenes per location (10 discs from each location)										Total, pcs	Mean of 10 discs
	1	2	3	4	5	6	7	8	9	10		
	(10m)	(20m)	(30m)	(40m)	(50m)	(60m)	(70m)	(80m)	(90m)	(100m)		
1	26	23	26	10	9	5	0	3	5	4	111	11.1
2	32	30	31	8	8	4	4	0	0	4	121	12.1
3	18	21	41	6	11	4	3	2	6	0	112	11.2
4	24	16	22	11	4	0	0	2	5	3	87	8.7
5	27	18	10	8	0	3	2	4	3	0	75	7.5
6	30	23	11	10	11	6	2	0	0	0	93	9.3
7	16	20	14	14	0	3	6	0	0	2	75	7.5
8	10	16	23	10	10	0	4	3	5	0	81	8.1
9	18	17	10	3	6	2	0	0	5	0	61	6.1
10	23	20	8	6	2	2	0	0	7	1	69	7.2
Total:	**224**	**204**	**196**	**86**	**61**	**29**	**21**	**14**	**36**	**14**	885	8.85

Stinging by the imago is the most common at the connection point (coronula) of the tubular flower and the achene. At this point a vulnerable, poorly protected surface is formed, that lasts for a 2 to 3 days. Here the bug is able to penetrate into the internal substance quite easily with its powerful stinging/sucking mouth organ. However, greening of the contents (the seed) hardly ever occurs at this point – which problem may especially be a crucial "technical" problem with the striped, alimentary purpose hybrids during acceptance of export consignments –, contrary to the damage caused at the shoulder part of the achene, which mostly (in 70% to 80% of the cases) results in the greening of the seed (chlorophyll formation started by the effect of sunshine, which penetrates through the large injuries).

The adults carry on their maturation nourishment on the achene, and also on the axis inflorescentiae, the torus, and squame of the sunflower. At the reverse side of the disc, scars are formed of the injuries, which typically ulcerate, and later get suberized. Such wounds may facilitate the penetration of various harmful fungi (the *Rhizopus arrhizus* FISHER, in the first place).
In the course of our study, we could also observe an "unusual" method of laying eggs of the *S. equestris*. This species usually lays eggs in the soils. However, on 27 August 2001, some specimen gave up the "normal" way, and laid their eggs among the achenes, and in the area sheltered by the outermost row of achenes, the periclinium and by some squame, too. (This had also been observed on one occasion earlier, on 11 August 1991. This phenomenon, by all means, contributes new information to the biology of the *S. equestris*)

Our observations have also made it clear that the classification of the *S. equestris* as an "auxiliary pest" has significantly changed in the years past.. This species is playing a more and more dominant role among the plenty of pests of the sunflower.
Studies on germination biology carried out prior to, or concurrently with, our studies have proved, among others, that the damage caused by these plant bugs may be significant – though they do not attack the germs directly –, because various species of fungi (*Alternaria* sp., *Botrytis cinerea*, *etc*.) may accumulate in the suction area, that have well-known harmful effects on the germ (Horváth-Bujáki, 1991).

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Summary

SPILOSTETHUS [= LYGAEUS] EQUESTRIS L., (HETEROPTERA: LYGAEIDAE), A PEST OF SUNFLOWER

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Western European, and especially German-based multinational food processing companies tend to have "alimentary purpose" sunflower hybrids and varieties grown in Hungary. This "commercial" product is highly preferred as substitute of walnuts, or as filling in chocolate, bread, other bakery products, or baked into the top layer thereof.
In early 1990s German companies rejected several truckloads of export goods due to "greening of the seed in the achene (kernel)". Almost 90% of the rejected consignments originated from the sandy ridge of the region between the rivers Danube and Tisza, which is severely infected with the *Asclepias syriaca* L. weed. Field investigations revealed unambiguously that the greening of the seed in the achene had been caused by the (*Spilostethus [= Lygaeus] equestris* L.), that is a plant bug species. This species is able to cause extensive damages and discontinuities in the so-called shoulder part of the achene with its powerful stinging-sucking mouth organ. In the damaged area of intensive chlorophyll formation begins due to the solar effect, which is the clear cause of the greening of the seed in the achene.
In our investigations we studied the industrial purpose sunflower plantations in two neighbouring areas (Bácsalmás and Katymár) in respect of the damage caused by the *S. equestris*. While in Bácsalmás (that district was less infected with *A. syriaca*) we found a damage of decreasing intensity (8.85 damaged achenes per sunflower disc) as advancing towards the centre of the plantation, while in the area of Katymár (this area was severely infected with *A. syriaca*), this value was almost three times as high, reaching 31.71 damaged achenes per disc. Though the numbers are not very high in themselves – assuming that a well-built disc contains 1100 to 1200 seeds –, but the damage may result in significant fall of quality, or even prevent exporting.
Our extensive research into the causes of the greening of the sunflower seeds made it clear that the *S. equestris* has a significant influence on the contents of the seeds it has damaged: it increases the proportion of linoleic acid ($C_{18:2}$) by about 2.5%. That affects the lasting quality of the achene unfavourably (Horváth and Bujáki, 1991).
For this reason, a comprehensive study of the biology of the *S. equestris* became necessary for the developing possible protection methods. Such studies also represented the beginning of the investigation of any external or endogenous factors, that might obstruct the successful use of alimentary purpose sunflower hybrids or varieties in the food industry.

EFFECT OF SOME ORGANOPHOSPHATE INHIBITORS ON ACETYLCHOLINESTERASE IN *FORFICULA AURICULARIA* ADULTS (DERMAPTERA: FORFICULIDAE)

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The importance of biological control and IPM in the agricultural production seems to become greater and greater. However, the introduction and use of beneficial organisms as biocontrol agents is difficult in practice, because of their sensitivity to pesticides. Possible solutions can be the utilisation of pesticides which are harmless to biological agents (Hassan, 1989) or to find (collect/select) tolerant or resistant strains of natural enemies (Grafton-Cardwell and Hoy, 1985). These goals may be achieved only with the study of the effects of pesticides on beneficial species.
Our aim in the present study was to analyze the detrimental effects of some organophosphorous insecticides on acetylcholinesterase(s) (AChE) of adult common earwig, *Forficula auricularia* L., an efficient polyphagous predator which feeds on aphids, codling moth, and scales (Helsen *et al.,* 1998, Schruft *et al.,* 1995). In addition, this analysis may contribute to the better knowledge of acetylcholinesterase(s) of the earwigs barely studied from this point of view.

Materials and Methods

Insects
F. auricularia adults derive from a field population collected in the experimental area of the Gembloux Agricultural University (Gembloux, Belgium) in 1995.
Captures were obtained by sweeping net. Insects were used for analyses immediately after captures.

Insecticides
All insecticide active ingredients were purchased: paraoxon, malaoxon, carbaryl, diazinon, schradran (Riedel-de Haën, Belgium).

Inhibition experiments

Preparation of homogenates

20 adult earwigs were decapitated then their heads homogenized with 2 ml cold HST buffer (Tris HCl 1 mM, pH 8; NaCl 1 M; Triton X-100 1% wt/vol; $MnCl_2$ 1 mM; $CaCl_2$ 1mM) in a motor-driven, Teflon pestle and glass tube tissue grinder. This homogenate was filtered through a thin layer of glass wool and centrifuged 15 min at 15300 g and 4 EC. Immediately after centrifugation the supernatant was used for AChE assay.

AChE assay

AchE activity in homogenates of earwig heads was estimated with the procedure of Ellman *et al.,* (1961) using a Shimadzu UV-160A spectrophotometer. The assay medium consisted of 10 µl of the following solution (36.6 mg DTNB (5-5-dithiobis 2-nitrobensoic acid) and 15 mg NaHCO3 in 10 ml phosphate buffer (0.1 M, pH 7.2)), sufficient Tris HCl buffer (1mM, pH 8) to give a final reaction volume of 1 ml, 10 µl acetone solution of paraoxon or malaoxon, 80 µl supernatant, 10 µl acetylthiocholine iodide (100 mM). Acetylthiocholine iodide was added only after a preincubation for 1 min to the other reaction components being in a cuvette. Then it was mixed rapidly and the change in absorbance was measured continuously at 412 nm for 1 min. Each enzyme assay included 9-15 cuvettes (two containing untreated (control) homogenates, 5-11 treated homogenates and two blanks). The blanks contained the same components except that substrate was omitted and instead of the pesticide acetone was added. At least duplicate analyses at each of 5-10 concentrations of pesticides were carried out.

Surface contact treatment

Chemicals

Parathion (range of 0.00028, 0.00083, 0.0025, 0.0075, 0.03, 0.12 %); the acetone solution of the active ingredients (300 µl/disc) was applied on Whatman paper discs. After the solvent evaporated, the disc was placed into a plastic petri dish with 8 cm diameter. 10 adults were immobilized to facilitate handling by exposing them briefly to carbon dioxide and then placed in each dish. There were 2 dishes per concentration. The test animals remained in the dish until a stable mortality resulted. The number of paralyzed or dead individuals was recorded after 1, 2, 4, hours, 1, 2,...days. Data were analyzed by probit analysis (Finney, 1971) with a program that incorporates Abbot's (1925) correction for natural mortality. All tests were conducted in the laboratory at 22-25 EC, 50-70 % RH, and under a 15:9 (L:D) photoperiod.

Results and Discussion

One population sample of *F. auricularia* was tested against the following classical AChE inhibiting insecticides: paraoxon, malaoxon, carbaryl, diazinon and schradran. Schradran showed the slightest efficiency. It was so small that it was not possible to determine its I_{50} value. As an approximate value it can be indicated that the I_{50} value must be considerably over 10^{-3} M which is the highest usable concentration. The other inhibitors' I_{50} and K_i values (Table 5, 6), except that of paraoxon, were - regarding its order of magnitude - very similar to those assessed on ladybird (*Coccinella septempunctata*) and lacewing (*Chrysoperla carnea* s.l. (Bozsik *et al.,* 1996). As to the paraoxon, earwig's AChE responded extremely susceptibly to it. Its I_{50} and K_i values represented extraordinarily higher efficiency than the corresponding values measured on ladybird and lacewing.
The pesticide active ingredients' efficiency order against the earwig AChE was the following: paraoxon > carbaryl > malaoxon > diazinon > schradran.
Comparing the efficiency of the surface contact effect of parathion on earwig with that on *C. septempunctata* (Bozsik et al., 1996), the earwigs were much more tolerant to parathion than the ladybirds (Table 3).

Table 1. Susceptibility of AChE of *Forficula auricularia* Adults from the Experimental Orchard of the Gembloux Agricultural University (Gembloux) to Paraoxon and Malaoxon
Abbreviations: Ge: Gembloux, Belgium; 95: 1995; n. e.: not examined)

Earwig	*Paraoxon*		*Malaoxon*	
Population	I_{50} (M)	K_i (M^{-1} min^{-1})	I_{50} (M)	K_i (M^{-1} min^{-1})
Ge95	3.6×10^{-6}	193188.3	5.5×10^{-6}	126086.3

Table 2. Susceptibility of AChE of *Forficula auricularia* adults from the Experimental Orchard of the Gembloux Agricultural University (Gembloux) to Diazinon and Carbaryl (Abbreviations see Table 1)

Earwig	Diazinon		Carbaryl	
Population	I_{50} (M)	K_i (M^{-1} min^{-1})	I_{50} (M)	K_i (M^{-1} min^{-1})
Ge95	3.4×10^{-3}	206.7	4.26×10^{-6}	163175.3

Table 3. Surface Contact Effect of Parathion on *Forficula auricularia* adults (Gembloux)

Time of evaluation (h)	LC_{50} (%) (95% FL)
72	0.1428
	0.0709 – 1.1366
96	0.1313
	0.0647 – 0.9022
144	0.0293
	0.0176 – 0.0542

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Summary

EFFECT OF SOME ORGANOPHOSPHATE INHIBITORS ON ACETYLCHOLINESTERASE IN *FORFICULA AURICULARIA* ADULTS (DERMAPTERA: FORFICULIDAE)

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In vitro enzyme activity of head homogenates of *Forficula auricularia* collected from the experimental area of the Gembloux Agricultural University and *in vivo* surface contact treatments with organophosphorous active ingredients on the same species were investigated. The *in vitro* studies based on the Ellman method showed that the acetylcholinesterase (AChE) of the studied *F. auricularia* population sensitivity order to the inhibitors was as follows here: paraoxon, carbaryl, malaoxon, diazinon, schradran. Comparing the I_{50} and K_i values with those of measured in *Cocccinella septempunctata* and *Chrysoperla carnea* s.l. it seems that the last two species are more tolerant to the AChE nhibitors acting *in vitro* then the earwig.

SPECIES SPECTRUM OF FLEA BEETLES (*PHYLLOTRETA* SPP., COLEOPTERA, CHRYSOMELIDAE) ATTRACTED TO ALLYL ISOTHIOCYANATE-BAITED TRAPS IN HUNGARY

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There are about 250 flea beetle spp. (Coleoptera, Chrysomelidae, Halticinae) present in the Carpathian basin, of which first of all those species cause agricultural camages, which live on cruciferous plants (mainly *Phyllotreta* spp.). In case of univoltine species most important damages are caused in the spring by the overwintering adults, on the one hand by feeding on leaves of seedlings, and on the other by propagating plant pathogens.
In the control of flea beetles the application of a trap would be very useful, which could detect the occurrence of overwintering adults in early spring, could monitor their flight pattern and could give estimates of the size of the populations of the overwintering generation, and also of the next generation of adults which appear later in the summer.
It has long been known that some plant volatiles, i.e. in the case of cruciferous plants isothiocyanates, which derive as secondary metabolites from the decomposition of non-volatile glucosinolates (feeding and oviposition stimulants), play the role of food attractants in the case of certain *Phyllotreta* spp. The objective of our present study was to investigate the species spectrum of flea beetles which respond to allyl isothiocyanate (which has been described as one of the most potent attractants for certain flea beetles) in Hungary, so that the applicability of this attractant for plant protection purposes can be evaluated.
At all experimental sites our traps baited with allyl isothiocyanate caught large numbers of *Phyllotreta cruciferae* Goeze. Catches in baited traps were always significantly higher than in unbaited ones. Our results confirm earlier reports on the attractivity of this compound towards this species described earlier from other parts of Europe and from North America (Canada). This species is one of the most important pest flea beetles in Hungary.

The second most frequently recorded species captured was *P. vittula* Redtb. in our experiments. Traps with allyl isothiocyanate clearly caught more than unbatied ones showing a strong attraction by this compound. No previous reports on allyl isothiocyanate attracting this species has been published.
Regularly significantly more beetles were caught in baited traps from *P. procera* Redtb. as well. Attraction by this compound has not been published before for this species either.
In a test conducted at Pusztaszabolcs, in a rape field, baited traps caught sizeable numbers of *P. balcanica* Heikert while no beetle were captured in unbaited traps. Similar results were obtained in case of *P. nodicornis* Marsham, in another test at Nadap. No mention of allyl isothiocyanate attracting these spp. was found in previous literature
In the case of *P. undulata* Kutsch. although significantly more beetles were caught in baited traps than in unbaited ones in a test at Budakalász, which may be an indication for the attractivity of allyl isothiocyanate towards this species, however, due to the overall low numbers caught this should be confirmed in future tests.
Apart from *Phyllotreta* spp., specimens of the close relative *Psylliodes chrysocephala* L. were also captured in significanlty larger numbers in baited than in unbaited traps, indicating that allyl isothiocyanate may play a role in the chemical communication of also this species. Scientists from the UK have already reported that certain isothiocyanates evoked an electrophysiological response on the antennae of *P. chrysocephala*, however, to the best of our knowledge ours is the first report on the field activity of the compound.
In some tests sizeable numbers of *Chaetocnema concinna* Marsh. were also captured. In this case however there were no differences between the catches of baited and unbatied traps, showing that allyl isothiocyanate did not influence the behavior of this species. This is not suprrising, since the species does not feed on cruciferous plants.
As for relative abundance of these flea beetle species, in the catch by traps with allyl isothiocyanate ca. 80-90% of specimens belonged to *P. cruciferae*; 10-15% were *P. vittula*, or *P. procera*. Other species occured only in small percentages, depending on experimental site. It is surprising, that of such, important pest species like *P. atra* Fabr., *P. undulata* or *P. nemorum* L. only very low numbers occasionally were captured. Further studies are needed to decide whether this was caused by the fact that allyl isothiocyanate is not attractive towards these spp., or whether these species were present in very low population densities at the test sites. Results are summered up in the Table 1.

Table 1. Species spectrum of flea beetles (*Phyllotreta* spp.) attracted to allyl isothiocyanate baited traps

Species captured:	Attractive activity of allyl isothiocyanate:
P. cruciferae	has been known from literature; confirmed in this study
P. vittula	was discovered in this study
P. procera	was discovered in this study
P. balcanica	was discovered in this study
P. nodicornis	was discovered in this study
P. undulata	was discovered in this study; needs further confirmation
P. chrysocephala	was discovered in this study; electrophysiological activity has been known before
C. concinna	was not observed in our studies

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POSTER SESSION
PHYTOPATHOLOGICAL
GROUP

VERTICILLIUM WILT ON GROUNDNUTS – CAUSAL AGENT AND POSSIBILITIES FOR INTEGRATED DISEASE CONTROL

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Verticillium wilt is worldwide spread on a big number of cultivated and wild species (Kranz et al., 1978; Porter, 1984). Most common causal agents are *Verticillium dahliae* and *Verticillium albo-atrum*. They have more than 350 host plants that belong to 160 genera and 60 families. *Verticillium dahliae* is a major pathogen that cause tracheomycosis in tropics and temperate climate zones (Pegg, 1974) and attacks woody and park species, vegetable, forage and arable crops (Bender and Shoemaker, 1984). That fungus is pathogenic to more than 250 plants (Fravel, 1989).

For the first time Verticillium wilt on groundnuts have been reported in Asia in 1937 (cit. Porter et al., 1984). During the second half of 19th century the disease spread quickly and nowadays is found in all groundnuts growing areas (Purss, 1961; Hci, 1967; Frank and Krikun, 1969; Jackson and Durhan, 1969; Issac, 1967; Kranz et al., 1978; Porter et al., 1984). Depending on climate conditions, varieties grown and technologies applied, yield losses vary between 2 and 60% (cit. Porter et al., 1984).

Verticillium wilt is difficult to be controlled with pesticides because pathogens survive for a long time in soil. Results can be obtained only when complex measures are undertaken – technological, chemical, biological and breeding programmes.

Tjamos et al. (1991) and Tjamos and Vellias (1997) reported the possibility for applying antagonistic fungi against *Verticillium dahliae*. Soil colonization with *Talaromyces flavus* can suppress microsclerotia formation and soil infestation of *Verticillium dahliae* especially in root zone.

Research done by Solarska (1997) pointed out that soil incorporation of *Talaromyces flavus* and *Trichoderma viride* before sowing is an useful practice against Verticillium wilt and other soil pathogens. Moreover *Trichoderma viride* has stimulating effect on some crops.

Using *Talaromyces flavus* Kersten (1997) has prepared formulation for Verticillium wilt control and made greenhouse experiments with rape.

Aiming limitation of inoculum some authors (Evans, 1966; Purss, 1961; Morris et al., 1984) recommend well-organized technology practices, including proper irrigation, crop rotation, seed material free from infection, weed control, and introduction of resistant varieties and chemical control. Soil treatment with metyl bromide and chlorpikrine had been used to control

inoculum (Sinclair, 1967; Krikun and Frank, 1982). Fungicides as benzimidazole, oxichinolinsulphate, metyl tyophanate, Basamid granulate, etc. can also be applied (Nakov et al., 1999).
Most authors pointed out that breeding for resistance was the most prospective method (Morris et al., 1987; Agrios, 1988). Valencia type and Spanish varieties are susceptible to Verticillium wilt (Curtis and Durhan, 1969). Highly resistant forms are found in American type – Georgia bunch (Smith, 1960). Results from resistance breeding programmes are published by Subrahmanyam et al. (1992).
In Bulgaria till present there is no information available about Verticillium wilt on groundnuts and studies on methods for *Verticillium* control. The aim of present research is to identify causal agents, to test antagonistic fungi against *Verticillium dahliae*, and to study susceptibility of Bulgarian groundnut varieties and lines to the pathogen.

Materials and Methods

Research experiments have been done at the Department of Phytopathology, Agricultural University, Plovdiv in the period 1994-2002.
Causal agent have been isolated based on standard phytopathological methods, from cultivars Kalina, Sadovo 2609, Sadovo improved, Orpheus, Rositza, Velikan and lines 3170, and 3078^{b}. Pathogenicity of the strains have been proved by Koch postules – artificial inoculation of varieties Kalina, Sadovo improved, Orpheus, Rositza, Velikan and line 3170, in growth chamber. Symptoms have been reported after 30-35 days, based on the following scale:

0 healthy plants;
0.1 roots infected and mild leave chlorosis;
1 symptoms at stem base and 0.5-1 cm above (vein necrosis); leaf chlorosis;
2 symptoms on root and stem (vein necrosis), chlorotic leaves and beginning of leaf necrosis.

Morphology and cultural characteristics of the colonies and mycelia (conidiophores, conidia) have been studied under microscope from 12-14 days culture on PDA.
Following cultivars have been inoculated with *Verticillium dahliae*: Sadovo 2609. Orpheus 3351, Kalina, Rositza 3092, Velikan, Sadovo 2510, Sadovo improved and lines 3078^{b}, 3371, 3403, 3301, 3675, 3202^{b}, 3256, 3240^{a}, 3170, 3190^{b}, 3301^{3} respectively. Experiments have been carried in sterile soil, in growth chamber, two series (10 plants each). Inoculation has been done with spore suspension when young plants appeared and at 2-4 true-leaf stage.

Results have been reported on 30^{es} and 50^{es} days after inoculation, based on the following grading scale:

1 Resistant – R – No symptoms;
2 Moderately resistant – MR – root tips are infected, mild chlorosis on older leaves;
3 Moderately susceptible – MS – roots and stem-base infected, chlorosis on older leaves;
4 Susceptible – S – roots, stem-base and 1.5-2 cm of the stem infected, beginning of leave necrosis.

Antagonistic activity of *Talaromyces flavus* and *Trichoderma viride* has been studied *in vitro*. Based on diameter of the colonies of pathogen and antagonists, coefficient of competition is calculated:

$$K = \frac{Da}{Df}$$

K – coefficient of competition;

Da – colony diameter of antagonist

Df – colony diameter of pathogen

Strains divide in 3 groups:

5 $K < 1$ – non competitive
6 $1 < K < 2$ – weak competitiveness
7 $K > 2$ – highly competitive

Other strains divided as follows:

8 Non active – $K < 1$, no antibiotic activity zone, no hyperparasitic reaction
9 Weak antagonists – $1 < K < 2$, sterile zone and/or hyperparasitism
10 Strong antagonists – $K > 2$, sterile zone and/or hyperparasitism

Results and Discussion

In our experiments 8 strains have been isolated from different varieties: Kalina, Sadovo 2609; Sadovo improved, Orpheus, Rositza, Velikan, lines 3170, 3078^{b}. Their pathogenicity have been proved and morphology studied on PDA. Colonies are whitish, fluffy, and rosy at the base. Mycelia are hyaline, with thin walls. Conidiophores formed on horizontal hyphae and vertically branched, short and hyaline. Rarely secondary branches are formed. Conidia are hyaline, single celled, elliptical, rarely ovoid, their size 3.15-7.10 x 1.2-3.4 μm. They are formed at the tip of conidiophores. On nutrient media microsclerotia are also formed. Data correspond with those published by Meloun and Wadsworth (cit. Porter, 1984).

Analysis of data and comparison with literature sources, lead to conclusion that in Bulgaria causal agent of Verticillium wilt on groundnuts is *Verticillium dahliae* Kleb. (Table 1).

Table 1. Conidia size of *Verticillium dahliae* and *Verticillium albo-atrum*, μm

Authors	*Verticillium dahliae*	*Verticillium albo-atrum*
Pidoplichko, N.M.	3-3,5 x 1,5-2	3-12 x 2,5-3
Christov, Al.	3,25-7 x 2-2,75	3,25-7 x 2-2,75
Chohriakov, M.K.	3-5,5 x 1,5-2	2,8-12 x 1,5-3
Melouk and Wadsworth	3 x 6.5	
Ibitui, Nakova	3.15 – 7.10 x 1.2-3.4	

Results from studies on antagonistic activity of *Talaromyces flavus* and *Trichoderma viride* are present of Table 2.

Table 2. Antagonistic activity of *Talaromyces flavus* and *Trichoderma viride* against different strains of *Verticillium dahliae*

Origin of isolate	Pathogen colonia diameter, mm	Antagonist colonia diameter, mm	Coefficient of competition, K
Bulgaria	*Verticillium dahliae* /groundnuts/ 47	*Trichoderma viride* - 69.5	1.47
Greece	*Verticillium dahliae* /olive/ 10	*Trichoderma viride* - 77.5	7.75 hyperparasitism
Bulgaria	*Verticillium dahliae* /groundnuts/ 50.5	*Talaromyces flavus* - 67.5	1.33
Greece	*Verticillium dahliae* /olive/ 20	*Talaromyces flavus* - 50	2.5

Trichoderma viride have weak antagonistic activity to *Verticillium dahliae* isolated from groundnuts (K = 1.47) and high competitiveness to the strain from olives /K = 7.75/.

Talaromyces flavus also express high antagonistic activity to *Verticillium dahliae*, isolated from olives (K=2.5).
Experiment data concerning cultivar response to *Verticillium dahliae* are present on Table 3.

Table 3. Response of some groundnut varieties and lines to *Verticillium dahliae*

Variety/line	*30es day*	*50es day*
Line 3675	R	MR
Line 3170	MR	MS
Line 3202[b]	MR	MS
Kalina	R	MR
Line 3190[b]	R	R
Sadovo 2609	MR	MS
Line 3256	MR	MS
Rositza 3092	MS	S
Line 3240[a]	MS	S
Orpheus	MR	MR
Line 3078[b]	MS	S
Sadovo 2510	MS	S
Velikan	MR	MS
Sadovo improved	MS	S
Line 3371	MR	S
Line 3403	MR	MS
Line 3301[3]	MS	S

On the 30es day after inoculation resistance is expressed by cultivar Kalina and lines 3675, 3190[b]; moderate resistance – from lines 3170, 3202[b], 3256, 3371, 3403 and varieties Sadovo 2609, Orpheus, and Velikan.
On the 50es day after inoculation resistant response has been observed only on line 3190[b]. Varieties Kalina and Orpheus, and line 3675 showed moderate resistance.
Based on studies and results obtained following conclusions can be made:

- As a causal agent of Verticillium wilt on groundnuts in Bulgaria *Verticillium dahliae* Klebahn has been isolated and identified.
- *Trichoderma viride* and *Talaromyces flavus* have weak antagonistic activity to *Verticillium dahliae* isolated from groundnuts and high competitiveness to the strain from olives.

- From cultivars tested for resistance to *Verticillium dahliae* on the 50^{es} day after inoculation only line 3190^{b} showed resistance, and Kalina, Orpheus and line 3675 were moderately resistant.

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Summary

VERTICILLIUM WILT ON GROUNDNUTS – CAUSAL AGENT AND ALTERNATIVE POSSIBILITIES FOR CONTROL

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Verticillium wilt is a disease of economic importance and can influence the yields in groundnut production. Our goal has been to identify causal agent and study biological means (antagonistic fungi) for control, as well as to test varietal resistance of some groundnut cultivars and lines.
As a causal agent of Verticillium wilt on groundnuts in Bulgaria *Verticillium dahliae* has been isolated and identified. The pathogen has hyaline mycelia and simple conidiophores, vertically branched rarely with secondary branches. Spores formed at the top of conidiophores are single celled, elliptical, hyaline, size 3.15-7.10 x 1.5-2.2 μm.
Talaromyces flavus and *Trichoderma viridae* have high antagonistic activity to *Verticillium dahliae* isolate from olives, but not to *Verticillium dahliae* from groundnuts.
From cultivars and lines tested for resistance to *Verticillium dahliae* the line 3190^{b} showed resistance, line 3675, varieties Kalina and Orpheus were moderately resistant after the 50es days.

RESISTANCE OF WINTER WHEAT CULTIVARS AGAINST NECROTROPHIC LEAF PATHOGENS
(2001-2003 SZEGED, HUNGARY AND 2003 ASCHERSLEBEN, GERMANY)

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The infections by *Drechslera tritici-repentis*, *Septoria tritici*, *Stagonospora* (*Septoria) nodorum* and *Bipolaris sorokiniana* significantly increased due to the poor economic situation, increasing monocultural and minimum tillage practices in Hungary at the late '90s. Medium epidemic was observed in 1996 and 1999 in the Szeged nurseries caused by *S. tritici*. In 1996, Pál Békési recorded in Kecskemét a medium level of epidemic. The two data series often showed significant differences for the same cultivars.
D. tritici-repentis was described first in Hungary by Aponyi et al. (1988). The available data basis is poor, mostly data about influence on monocultural production were reported (Balogh et al. 1991, Rátai and Pecze 1997).
We have not enough information about the resistance of winter wheat cultivars that is why a new project studying resistance and yield reaction of the varieties has started.

Materials and Methods

1.) ***Testing of cultivars after winter wheat:***
Thirty-two registered cultivars were tested during three years in Szeged. The plot size was $4m^2$, in four replicates, under untreated and treated environments (randomized block design). The previous crop was winter wheat. Applied fungicides in 2001: Folicur Top (1 l/ha, tillering), Sphera (1 l/ha, at flowering); in 2002 and 2003: Juwel (1 l/ha) (at flowering, early spraying was omitted because occurrence only sporadic symptoms).
We scored the leaf spots, powdery mildew (*Blumeria graminis),* leaf rust *(Puccinia triticina)* and yellow rust (*Puccinia striiformis).* Leaf samples were collected from each cultivars. The samples were incubated in Petri dishes on wet filter-paper at 20 ^{o}C for 48-72 hours, and then microscopic identification of the necrotrophic fungi was performed (*D. tritici-repentis, S.*

tritici, S. nodorum and *B. sorokiniana).* After harvesting we measured the yield and the results were evaluated by two-way ANOVA.

2.) ***Testing of cultivars under artificial infection environment in adult and seedling stage***

The same thirty-two cultivars were tested in Aschersleben, in 2003. Ten *D. tritici-repentis* isolates collected from naturally infested wheat grown in Russia, Czech Republic and Germany were used for the field experiments and three from this for the tests in the greenhouse. The two rows of each wheat cultivars were separated from each other by one row of a susceptible cultivar and tested in the field. Infested oat kernels (a mixture of ten isolates) were used as method of inoculating wheat in the field. The oat kernels were dispensed in the field at the end of December. Tan spot was visually assessed on approximately 7-day intervals beginning 20 May 2003 using percentage of leaf attack.

The evaluation of resistance in the greenhouse was performed in 1-leaf seedling stage on detached leaves. Leaf segments were cut from each plant and arranged in trays on cotton soaked with 40 ppm benzimidazole solution (Mikhailova and Kvitko, 1970) and sprayed with conidia suspended in solution of Tween-80 at a concentration 6000 spores/ml. The reaction type was recorded 5-7 days after inoculation using a 0-5 scale (size of necrotic spots and necrotic halos) described by Lamari and Bernier,1989.

Results

Leaf rust epidemic was heavy, powdery mildew epidemic was medium during the last three years. We observed a medium yellow rust epidemic in 2001. Level of leaf spots epidemic was the highest in 2002 years (Table 1).

Table 1. Level of epidemic of the observed diseases

Years Observed diseases	2001	2002	2003
Leaf spots (%)	14.83	38.20	15.77
Powdery mildew (ACI)*	19.49	11.79	2.84
Leaf rust (ACI)	37.99	41.50	41.13
Yellow rust (ACI)	22,98	0,00	0.00

* average coefficient of infection (CIMMYT, 1976)

Leaf samples from each cultivar were collected and identified for the following necrotrophic pathogens in the leaf samples (Table 2).

Table 2. Identified necrotrophic pathogens (%)

Years Necrotrophic pathogens	2001	2002	2003
D. tritici-repentis	15.91	85.00	29.20
S. tritici	2.27	23.00	12.50
S. nodorum	13.64	2.00	12.50
B. sorokiniana	2.27	17.00	2.90

The occurrence of necrotrophic pathogens differed significantly even in the same location among years.

The mean yield response of the 32 cultivars (2001-2003) differed significantly between the protected and not protected experiments showing the general yield loss that was between 7.11-17.97%. Considering correlation coefficient the biotrophic pathogens influenced more expressed on the yield response of the cultivars (Table 3)

Table 3. Values of correlation coefficient (n=32)

Diseases	Years	Yield decrease %
Leaf spots	2001	- 0.2726
	2002	- 0.1999
	2003	0.1715
Blumeria graminis	2001	-0.6517***
	2002	-0.1545
	2003	-0.3350
Puccinia triticina	2001	-0.2423
	2002	-0.3730*
	2003	-0.1287
Puccinia striiformis	2001	-0.1318

On the base of both seedling and adult tests the most resistant cultivars were GK Héja, GK Selyemdur, GK Góbé, GK Holló, GK Margit, GK Bétadur, GK Favorit, and GK Hattyú. These possibly connected with having some resistance gene against *D. tritici-repentis*. Among these cultivars GK Holló and GK Margit were resistant both in seedling and adult stages. GK

Selyemdur was resistant in adult stage but susceptible in seedling stage. However GK Marcal was resistant in seedling stage and susceptible in adult one. It seems that these cultivars possess specific genes in adult and seedling stages. The clarification of the resistance background needs further investigations (Table 4).

Table 4. Resistance of winter wheat genotypes against *D. tritici-repentis* in adult and seedling stage (Aschersleben, Szeged)

	Leaf spots (%)						
Location	Aschersleben		Szeged	Aschersleben			
Cultivar	adult stage		average of three years	seedling stage			
	field	greenhouse	field	Kp1		Kp6	Kp12
GK Héja	10.0	R	10.0	R	R	MR	R-MR
GK Selyemdur	-	S	10.3	S	MS-S	S	MS
GK Góbé	40.0	S	15.4	S	MS	MS	MS
GK Holló	3.0	MR	15.7	-	R/MS	R	R
GK Margit	5.0	R	17.7	R	R	R	R
GK Bétadur	5.0	MR	18.3	MR-MS	MR	MR	MR
GK Favorit	10.0	R	19.6	-	R-MR	R	R
GK Hattyú	25.0	S	19.6	MR	R	R	R-MR
GK Marcal	35.0	S	37.5	R	R	R	R

R- resistant, MR- moderate resistant, MS- moderate susceptible, S- susceptible

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Summary

RESISTANCE OF WINTER WHEAT CULTIVARS AGAINST NECROTROPHIC LEAF PATHOGENS (2001-2003 SZEGED, HUNGARY AND 2003 ASCHERSLEBEN, GERMANY)

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Thirty-two winter wheat cultivars were tested in Szeged after winter wheat in protected and unprotected environment. Among the necrotrophic pathogens, the dominant pathogens were *Drechslera tritici-repentis* (Died.) Shoem. (2001, 2002 and 2003) and *Septoria nodorum* (2001). The resistance differences were significant. The biotrophic (leaf rust, yellow rust and powdery mildew) as well as the mentioned necrotrophic pathogens caused significant yield decrease in three years. According to values of correlation coefficients, the influence of biotrophic pathogens was greater on the yield.

Same cultivars were tested under artificial inoculated environment (the artificial inoculation was made by *D. tritici-repentis*) in the field and the greenhouse (in seedling stage, three isolates) in 2003, in Aschersleben, Germany.

Among the analyzed cultivars were not completely resistant against *D. tritici-repentis*. The data series often showed significant differences for the same cultivars. According to the above tests, the most resistant cultivars against leaf spots were **GK Héja, GK Holló** and **GK Margit**.

STUDY ON NATURAL ALTERNATIVES FOR THE CONTROL OF SUDDEN WILT INFESTING CANTALOUPE UNDER EGYPTIAN CONDITIONS

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Pathogenic soilborne fungi i.e. *Fusarium, Rhizoctonia, Pythium, Phoma* spp., etc. cause serious problem of sudden wilt (vine decline) are considered as dangerous organisms threatening vegetable production in the nursery, covered agriculture and open fields, because of the decrease in plants number and quality (Gwynne et al.,1997)

These pathogens of sudden wilt especially in some crops such as cantaloupe and watermelon cause rotting , pre and post-emergence damping off, while the older plants are affected by wilting or bad growth during flowering or fruiting stages reflecting on plant growth and on the yield quantity and quality (Martyn and miller 1996). The recommended systemic or contact chemical fungicides for the control of such pathogens are extremely harmful in the short or long terms on the man health and on the environment, causing dangerous diseases i.e. cancer, kidney, liver diseases and others (Mansour,1 992). Because of the importance of this subject we tried to do this applied research program for facing the cantaloupe sudden wilt problem in Egypt through finding safe alternatives for the control of such diseases, by avoiding the use of chemical fungicides or other materials such as methyl bromide for soil treatments in the control of these economic diseases by utilizing other non-chemical and safe means of natural origins such as: antagonistic fungi i.e. *Trichoderma* and *Glicoladium* (Cooney and Lauren.1998 and Charati et al., 988), and antagonistic bacteria i.e. *Bacillus, Streptomyces* spp., (Bochow,1989), plant materials i.e. Cinnamon, Garlic,Fenugreek, Rocket, onion and Camphor (Jiratko 1994 and El-sherbiny 2001), beside some new physical methods such as soil solarization in field trials against such soilborne cantaloupe pathogens (Bell et al., 1991)

Materials and Methods

Isolation and identification of used pathogenic or antagonistic fungi :

Two species of different fungal pathogens; *F. solani* and *F. oxysporum* f.sp. *melonis* were tested for its pathogenicity, all were isolated from root diseased seedlings of muskmelon obtained from fields at different locations at Dakahalia Governorate for laboratory tests on PDA. Fungal isolation from soil was done according to (Warcup 1950). Identification of pure

culture of each fungal pathogen was carried out through Dept.of plant pathology. Faculty of Agriculture and Faculty of Sciences at University of Mansoura, according to Gilman (1957), Barnett and Hunter (1972) stock culture of each fungal pathogen were kept on PDA slant for further studies.
Pure culture of antagonistic fungi *Trichoderma viride, Coniothyrium minitans* and *Glicoladium virens* were identified by Prof. Dr László Vajna Dept. Plant Pathology, Plant Protection Institute HAS, Budapest. Hungary, and Prof Dr. M. El-Sheshtawi, Dept. of Pant Pthology Faculty of Agriculture, Mansoura University, Egypt.

Laboratory Experiments

1 Inhibitory effect of certain fungal antagonists on radial growth of some soilborne fungal pathogens infesting cantaloupe
The inhibitory effects of *T. viride, G. virens* and *C. minitans* on radial growth of *F. solani* and *F. oxysporum* f.sp. *melonis* were studied. All pure cultures of the above mentioned 3 fungal antagonistic fungi, and the two fungal pathogens were grown on PDA for 5-7 days (25±2^{O}C). The antagonistic effect of the used antagonists on the fungal pathogens was done through using on disc (5 mm in diameter) of the antagonist facing one disc of the pathogen on the PDA surface and relatively closed to the periphery of the plates. The untreated control treatment was done on the same medium in Petri dishes by growing one disc of the pathogenic fungus in the center of medium surface according to (Johanson et al.,1959).
The *C. minitans* was grown on PDA for 7days and was placed in a side of the Petri dish and incubated at (25±2^{O}C) for 3 days then pathogenic fungal discs (5mm in diam.) were put in the other side of the dish in 5 replicates. All plates were incubated at (25±2^{O}C) for 7 days, then incubated for 4 days after inoculation, the diameter average of zones of the pathogenic fungus was recorded.

2 Effects of some crushed plant materials on radial growth of some soilborne fungal pathogens infesting cantaloupe
The inhibitory effects of some plant materials; Cinnamon, Garlic, Fenugreek, Rocket and Camphor on *F. solani,* and *F. oxysporum* f.sp. *melonis* were studied. Plant specimens were washed , dried and crushed by electric miller , treatments were of the doses zero, 2, 3 and 4 g/L of PDA, then autoclaved, 5 replicates were conducted, each replicate consisted of 5 Petri dishes of 9 cm diam., which were inoculated with 5mm diam. 6days old fungal discs, the concentration zero was free of any crushed plant material but inoculated with the tested fungal pathogens discs. All plates were incubated at (25±2^{O}C) for 7 days and the average radial growth was recorded and compared with control %.

3 Effect of some essential oils on radial growth of some soilborne fungal pathogens infesting cantaloupe

The three essential oils of Nigella, Onion and Camphor of 0.5, 1.0 and 1.5% concentrates were used with the two used fungal pathogens.The used PDA was supplied with 0.5% of tween 80 and with the essential oil concentrates before being solid (Katherine et al., 1998). Control treatment was done by mixing PDA with tween 80 only, and no essential oils were added, while treatments of various concentrates in Petri dishes were inoculated with the tested pathogens by giving on (5 mm diam.) fungal discs per each. Incubation was done for 7 days for *F. solani*, and *F. oxysporum* f.sp. *melonis* and average radial growth was recorded and compared with the untreated control %.

Greenhouse experiments

1 Phytopathogenicity tests

The test was carried out for studying the pathogenicity of the isolated soilborne fungal pathogens (*F. solani* and *F. oxysporum* f.sp. *melonis*) on 10 hybrids of Cantaloupe; Regal, Galia, C.8, Vicar, 1022, Caruso, Ideal, Mirella, Primal and Super VIP. Plastic pots of 25 cm diam. were filled with autoclaved sandy loam (50% sand+ 50% clay soil about 2 kg/ pot) then artificially infested with spore suspension of the pathogenic fungus (25 ml / pot) pathogenic fungi were cultured in 250 ml flasks containing 100 ml of potato dextrose broth for 7 days at (25 ± 2^{O}C). The mycelia in each flask were added to 200 ml of sterilized water in a satirized blender for 20 seconds at the low speed.The resulting suspension was used for soil artificial inoculation by 8 days. Irrigation took place immediately after planting, and repeated every 3 days during the duration of experiment .The planted pots were kept under the plastic house conditions during November and December where daily temperature average was (20 ± 2^{O}C), the experiments contained 3 replicates.

Data of disease incidence were recorded after 20 days of sowing for the pre-emergence damping-off and after 40days for wilting and compared with untreated and chemical controls.

2 Effect of Cantaloupe seed dressing with antagonistic fungi on the development of damping-off disease

Biological control trial in pots against damping-off disease caused by some soilborne fungal pathogens was conducted using fungal antagonists to study the effect of *T. viride, G. virens, C. minitans* in controlling damping-off disease of cantaloupe seedling caused by *Fusarium oxysporum* f. sp.

melonis using seed coating with conidial spores of used antagonists (*T. viride , G. virens* and *C. minitans*).

Plastic pots were filled with autoclaved sandy loam soil (about 2 kg/pot) and artificially inoculated cantaloupe seeds were planted in the artificially infested pots. Planting was carried out 8 days after inoculation, 3 seeds of the hybrids (Vicar, Primal, Ideal) were planted (3 replicates) at 1 cm depth under soil surface of each pot . Prior to planting, seeds were surface sterilized and coated with conidial spores of *T. viride, G. virens* and *C. minitans* (10 days old). Surface sterilization of seeds was carried out by dipping in 10% commercial hypochlorite sodium (Clorox) for 10 min.; washed through distilled water; then dried between sterile filter paper sheets. Seeds coating with fungal spores was performed by wetting them with sterile water containing molasses (as sticker), air dried and then placed on the surface of 6 days-old culture of *T. viride, G. virens* and *C. minitans* in Petri dishes in which conidia were abundant. Control treatment was done by soaking seeds in distilled water, while standard (control) chemical seed treatments were done by Topsin-M 70 1 g/kg and Thiram 2g/kg. Planting was done in all cases in artificially infested and non-infested soils. Data of damping-off and wilting was recorded after 21 and 40 days.

Irrigation took place immediately after planting and repeated after 3 days during the duration of experiment .The planted pots were kept under the plastic house conditions during November and December where daily temperature average range ($20\pm2^{O}C$). The experiment contained 3 replicates for each treatment .Data of disease incidence were recorded after 20 days of sowing and after 40 days for and compared with the four controls (mentioned above).

3 Effect of cantaloupe seed soaking in plant materials on the development of damping-off disease

Discs of (5mm diam.) were taken from 7 days old soilborne fungal pathogen cultures (mentioned in abstract) were transferred to PD broth (as mentioned before). Healthy seeds of the cantaloupe hybrids (primal, Ideal, vicar) were soaked for 4 hours in cinnamon and Eucalyptus material at concentration of 75%. Treatments were as follows:

Soaking in Cinnamon and Eucalyptus materials 4 hours, control treatment was done by soaking in distilled water for 4 hour, while chemical seed treatments were done by Topsin 1 g/kg and Thiram 2g/kg. Planting was done in all cases in artificially infested and non-infested soils. Data of damping-off and wilting were recorded after 20 and 40 days.

4 Effect of Cantaloupe seed soaking in essential oils on the development of damping-off disease

After the artificial inoculation, of soil in pots, healthy seeds of Cantaloupe hybrids (Vicar, Primal, Ideal) were soaked seperatly for 30 min. in each oil of Nigella, Onion and Eucalyptus at concentrate of 0.5%. Control treatments were done.

5 Effect of biofertilizers on the development of damping-off disease:

Trials were carried out to study the effectiveness of biofertilizers (Microbin, phosphorin and Rizobacterin) on the examind two soilborne pathogens (mentioned in the abstract) Biofertilizers were mixed with artificially infested soil in pots in the upper 10 cm, then seeds were planted in infested pots as previously mentioned, taking in consideration that the standard chemical fungicide Topsin-M70 was used as soil drenching with concentration of 1.0 %,while the Thiram concentration was 0.2 %, each 25 cm diameter pot received 25 cm^3.

6 Soil solarization and methyl bromide under field condition

Two trails are designed to be done through cooperation with the Agriculture Research center (A.R.C.), Dept. of Covered Agriculture, Vegetable Section of the Institute of Horticulture.

The first trails is done already at Ismailia, (East bank of Suez canal season of 2000/2001) Data is over, while the other trail is done at Aswan (season 2001/2002), Data in print.

The trail of 2000-2001 at Ismailia included the following treatments:

1 - Soil Fumigation with methyl bromide at the rate of 35 g/m^2

1 a - Soil Fumigation with methyl bromide at the rate of 35 g/m^2 + soil solarization

2 - Soil Fumigation with methyl bromide at the rate of 70 g/m^2

3 - Soil solarization alone

4 - Untreated soil (control)

Methyl bromide applications are done before seed sowing with one week, soil solarization was done for nine weeks during June, July, and August 2000. All treatments of this trail were arranged in complete randomized block design with three replicates ,each replicate contained 60 plants .Other Agriculture treatments were applied as recommended by A.R.C, - H.R.I Protected Cultivation Department. Mineral fertilizers were applied through drip irrigation system the hybrid primal from Holland was used in this experiment.

The second trail of 2001/2002 conducted at Aswan (1000 km far from Cairo) was done by soil solarization without any chemicals, trail is still repeated till the end of 2003.

Statistical analysis

Statistical analyses of all experimental data of this field trail were done using the statistical software package Costat (1990).

All comparisons were first subjected to one way ANOVA and significant differences between treatment means were determinate.

Results and Discussion

Laboratory experiments

Effect of certain antagonistic fungi on radial growth of some soilborne fungal pathogen infesting cantaloupe

Among all tested antagonistic fungi, *T. viride* gave the best inhibitory effect between all tested soilborne pathogenic fungi; this antagonist reduced the radial growth of *F. oxysporum* f.sp. *melonis* by 70.4 % when compared with controls (Table 1). These results agree with some authors who reported that *T. viride* was able to suppress the growth of one or more of these fungi (Zhao- Gouai, *et al;* 1998; Thiribhuvanamala, *et al;* 1999; prodeep, *et al;* 2000 and Mathivaran *et al;* 2000). They found that *T.viride* eliminated *R. solani*, *F. oxysporum* f.sp. *niveum*, *F. solani* and *A. alternata*. This high antifungal activity of *T. viride* is possibly due to some enzymes and secondary metabolites (Antal, et *al;* 2000) who reported that the activities of extra cellular chitinase (EC 3.2.-1-30), Beta-glycosidase (EC3-4-21; EC3-4-21-4), which are thought to be involved in the mycoparasitic process. In addition, (Cooney and Lauren, 1998), found that the antifungal *Trichoderma* secondary metabolite 6-n-pentyl-2-H-pyran-2-one level significantly increased in the presence of the pathogen.

In our study, we noticed that *G. virens* gave moderate inhibition to mycelial growth of *F. solani,* and *F. oxysporum* f.sp. *melonis* with radial growth reduction rates of 33.5, and 60% respectively. These results agree with (Aghnoom, *et al*1999; Rispoli and Nicoletti1999; Prakash *et al*1999; Khan and Akram2000; Singh and Mukhopodhyay2000), they reported that *G. virens* inhibited the mycelial growth of *F. oxysporum, S. rolfsii* and *R. solani, C. minitans* slightly reduced the mycelial growth of *F. oxysporum* f.sp. *melonis* and *F. solani* with degrees of 47.3 and 26.4 % respectively when compared with control.

Table 1. Effect of some antagonistic fungi on radial growth of some soilborne-fungal pathogens

F. solani		*F. oxysporum*		Fungi
Inh%	R.G	Inh%	R.G	
62.4	24.4	70.4	26.6	*T.viride*
33.5	43.2	60.0	36.0	*G.virens*
26.4	47.8	47.3	47.4	*C.minitans*
0.0	65.0	0.0	90.0	Control

L.S.D 0.05= 1.00 0.01= 1.66
R.G = Radial Growth Inh% = inhibition percentage

Effects of some plant materials on radial growth of some soilborne fungal pathogens

Data in Table 2 revealed that, the crushed of Cinnamon material proved to be the most effective on radial growth for the tested fungi, suppressing radial growth of *F. solani* and *F. oxysporum* f.sp. *melonis* by 42.1 and 50% at the concentration of 4 g/l respectively. These results agree with some authors who reported the antifungal activity of cinnamon material against one or more of these fungi (Sinha, et al., 1993; Jiartko,1994; Michail, et al., 1994 and Scholz 1999), They reported that cinnamon material and its oil inhibited the mycelial growth and spore *germination* of *Fusarium* spp., *Alternaria* spp., *Cladosporium* sp., *P. italicum, A. niger* and *B. cinerea.*

This high antifungal activity of cinnamon is possibly belonging to some aldehydes and acid compounds as many authors reported (El- Maraghy 1995 and Wilson *et al.* 1997). They reported that cinnamon material contains cinnameldehyde, cinnamic acid, Beta- myrcene and Alpha- pinene compounds.

The present study revealed that the Garlic material effect came on the second class against the mycelial growth of the tested fungal soilborne pathogens, these results are in agreement with (Gupta and Sharma, 1993; Karade and Sawant, 1999; Sharma and Kapoor, 1999 and Brown, et al., 2000), who reported that garlic material inhibited the mycelial growth of *A. alternata, S. sclerotiorum, M. phaseolina, F. solani, R. solani* and *P. italicum*, also inhibited spore germination of *F. solani* and *A. alternata*, they also mention that this material contains ((E.2)-4,4,9- trithiadodeca 1,6 11-triene-9-oxide), a derivative of allicin.

Table 2. Effect of some plant materials and some essential oils on radial growth of *F. oxysporum* f.sp. *melonis* and *F. solani* infesting cantaloupe

F. solani		*F.oxysporum* f.sp. *melonis*		Conc.	Plant sources
Inh%	**R.G**	**Inh%**	**R.G**		
18.6	37.2	38.8	55.0	**2g/L**	***C. zeylanicum***
28.3	64.5	44.1	50.2	**3g/L**	
42.1	52.0	50.0	45.0	**4g/L**	
9.4	81.4	14.7	76.8	**2g/L**	***T. foenum***
16.5	75.2	32.1	60.9	**3g/L**	
25.2	67.3	33.2	60.1	**4g/L**	
12.8	78.4	30.1	62.9	**2g/L**	***E. globulus***
21.7	70.4	34.1	59.2	**3g/L**	
29.2	63.7	41.5	52.6	**4g/L**	
5.8	84.7	20.4	71.6	**2g/L**	***E. sativa***
11.7	81.4	33.2	60.1	**3g/L**	
19.3	72.6	38.8	55.0	**4g/L**	
13.5	77.8	30.5	62.2	**2g/L**	***A. sativum***
21.0	71.1	33.8	59.5	**3g/L**	
32.8	60.4	43.0	51.3	**4g/L**	
	2.00 2.66		N.S N.S	**L. S. D. 0.05 - 0.01**	
				Conc.	**Essential oils**
78.6	19.2	88.0	10.7	**0.5%**	***A. cepa***
88.7	10.0	100.0	0.0	**1.0%**	
100.0	0.0	100.0	0.0	**1.5%**	
17.7	74.0	2.6	87.6	**0.5%**	***N. sativa***
21.1	71.0	23.1	69.2	**1.0%**	
37.1	56.6	34.7	58.8	**1.5%**	
32.0	60.8	39.5	54.4	**0.5%**	***E. globulus***
100.0	0.0	63.0	32.6	**1.0%**	
100.0	0.0	78.3	19.5	**1.5%**	
0.0	90.0	0.0	90.0	**Control**	
	0.35 0.56		0.33 0.53	**L. S. D. 0.05 0.01**	

Conc.= Concentration, R.G. = Radial Growth, Inh.% = inhibition percentage
N.S. = Non significant

It is noticed also that some plant materials slightly suppressed the fungal growth of the tested pathogens. For example, Eucalyptus on *F. solani* gave (29.2%) mycelial growth reduction.

Inhibitory effect of some essential oils on radial growth of some soilborne fungal pathogens

Among all tested essential oils, onion oil mixed with PDA at concentrate of 1.5% proved to be the most effective growth inhibitor for *F. solani*, *F. oxysporum* f.sp. *melonis*, giving 100% reduction in both cases (Table 2),. These results agree with (Favaron, et al., 1993; Zohir, et al., 1995; Wilson, et al., 1997, Fan and Chen, 1998, 1999), who observed antifungal activity of onion oil against the mycelial growth and spore germination of *F. moniliforme, B. cinerea, S. cepivorum* and *S. sclerotiorum*. Results obtained by (O'Gara et al., 2000) show that onion oil contains some sulfur compounds and he mentioned that oil had antifungal and antibacterial activities.

The present study revealed that oil of *Eucalyptus* gave similar results to Onion against *F. solani*, when it reduced the fungal growth of these fungi by 100%, this oil gave also 78.3% inhibition of the mycelial growth of *F. oxysporum.* f.sp. *melonis*, such results agree with (Raghavaiah and Jayaramaiah, 1987; Singh and Dwivedi, 1990; Ansariand Shrivastava, 1991, Singh and Gupta, 1992), who found that *Eucalyptus* oil inhibited the linear growth of *Cercospora* sp., *F. solani, F. moniliforme, R. solani* and *S. sclerotiorum.*

In our study, *Nigella* oil had moderate inhibitory effect on the radial growth of *F. solani*, *F. oxysporum* f.sp. *melonis* by 37.1 and 34.7% at 1.5%, respectively. These results agree with (Rathee et al., 1983; El-Kayati et al., 1995 and Rahhal 1997), who found that the essential oil of *Nigella (N. sativa)* was effective against *R. solani*, *F. solani*, *F. oxysporum, S. sclerotiorum, Pythium vexans* and *F. moniliforme*.

In general we observed from our tests on the inhibitory effect of various tested plant oils on the 5 tested soilborne fungal pathogens that the best effect came from Onion oil followed by *Eucalyptus* oil and then the *Nigella* oil. About *Eucalyptus (E. globulus)* we noticed that when comparing the inhibitory effect of its material and its oil on all tested pathogens that both gave various inhibitory values.

Greenhouse experiments

Effect of antagonistic fungi on the development of the damping-off disease infesting 3 cantaloupe hybrids

Results in (Table 3 and 4) showed that, seed treatment with *T. viride* and *G. virens* gave good inhibitory effects of 88.89% protection against the damping-off caused by *F. oxysporum* f.sp. *melonis* and *F. solani* when living control plants percentage was 44.4%, while seed treatment with fungicides (Thiram and Topsin), gave 88.89% and 100% disease control respectively obtained by (Goundar and Srikant, 1999, Hoda-Ahamed et al.,

2000; Rajapan and Yesuraja, 2000, Dubey, 2000), who reported that *T. viride* gave good result when compared with Captan, Vitavax and Carboxin.

Table 3. Effect of seed dressing with some bioagents on the disease incidence on cantaloupe hybrids caused by *F. oxysporum* f.sp. *melonis.*

Ideal					Primal					Vicar					Treatments
Survival %	B		A		Survival %	B		A		Survival %	B		A		
	M%	N0	M%	N0		M%	N0	M%	N0		M%	N0	M%	N0	
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Non – infested
44.45	22.22	1.33	33.33	2.00	55.56	11.11	1.66	33.33	2.00	44.45	11.11	1.33	44.44	1.66	(Infested)
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	Thiram
100	0.00	3.00	0.00	3.00	0.00	0.00	3.00	3.00	3.00	100	0.00	3.00	0.00	3.00	Topsin
88.89	11.11	2.66	0.00	3.00	88.89	0.00	2.66	11.11	2.66	88.89	0.00	2.66	11.11	2.66	*T. viride*
88.89	0.00	2.66	11.11	2.66	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	*G. virens*
77.78	11.11	2.33	11.11	2.66	77.78	22.22	2.33	0.00	3.00	77.78	11.11	2.33	11.11	2.66	*C. minitans*
L.S.D	0.05 =			0.22					0.41					0.25	
		0.37					0.66					0.01 =			0.36
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	0.00	2.66	11.11	2.66	*C. zeylanicum*
77.78	22.22	2.33	0.00	3.00	88.89	0.00	2.66	11.11	2.66	66.67	0.00	2.00	33.33	2.00	*E. globulus*
L.S.D	0.05 =				0.24					0.43				0.23	
		0.71				0.69					0.01 =				0.38
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	*A. cepa*
77.78	11.11	2.33	11.11	2.66	77.79	0.00	2.33	22.22	2.33	77.78	11.11	2.33	11.11	2.66	*N. sativa*
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	*E. globulus*
L.S.D	0.05 =			0.22			0.42				0 .75				
			1.22					0.68				0.01 =			0.35

NO= Number of plants M % = Mortality percentage
A= After 20 days from planting B = After 40 days from planting
* Vicar : slightly tolerant, * Primal : highly tolerant, * Ideal : moderately tolerant

Effect of some plant materials on the development of the damping-off disease infesting 3 cantaloupe hybrids

Data in (Tables 3 and 4) show that the effect of plant materials in controlling Cantaloupe damping-off under greenhouse was relatively high giving 88.9 -100 protection in case of *Cinnamon (C. zeylanicum). Eucalyptus (E. globulus)* material gave 77.8% protection, while chemical treatment with both Topsin-M and Thiram gave 100% and 88.89 % protection respectively when compared with untreated controls gave living plants percentages ranged between 44.4% and 55.5%.

Table 4. Effect of seed dressing with some bioagents on the disease incidence on Cantaloupe hybrids caused by *F. solani*

Ideal					Primal					Vicar					Treatments
Survival %	B		A		Survival %	B		A		Survival %	B		A		
	M%	N0	M%	N0		M%	N0	M%	N0		M%	N0	M%	N0	
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Non –infested
44.45	22.22	1.33	33.33	2.00	55.56	11.11	1.66	33.33	2.00	22.23	33.33	0.66	44.44	1.66	(Infested)
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	Thiram
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Topsin
88.89	0.00	2.66	11.11	2.66	88.89	0.00	2.66	11.11	2.66	88.89	11.11	2.66	0.00	3.00	*T. viride*
88.89	0.00	2.66	11.11	2.66	88.89	0.00	2.66	11.11	2.66	77.89	11.11	2.33	11.11	2.66	*G. virens*
77.78	11.11	2.33	11.11	2.66	88.89	11.11	2.66	0.00	3.00	55.56	33.33	1.66	11.11	2.66	*C. minitans*
L.S.D 0.05 =			0.24					0.43					0.23		
88.89	0.00	2.66	11.11	2.66	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	*C. zeylanicum*
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	*E. globulus*
L.S.D 0.05 =			0.24					0.43					0.23		
	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	88.89	0.00	2.66	11.11	2.66	*A. cepa*
88.89	0.00	2.66	11.11	2.66	88.89	11.11	2.66	0.00	3.00	66.67	11.11	2.00	22.22	2.33	*N. sativa*
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	*E. globulus*

NO= Number of plants M % = Mortality percentage A= After 20 days from planting B = After 40 days from planting

Effect of some essential oils as seed disinfectant materials on the development of the damping-off disease infesting 3 Cantaloupe hybrids

Effect of both Eucalyptus *(E. globulus)*and Onion *(A. cepa)* essential oils gave the highest level of control to damping-off disease giving of cantaloupe plants (100%) followed by Nigella *(N.sativa)* which gave 88.89% protection, while control was (44.4%) (Tables 3 and 4), They gave the good results when compared with the chemical fungicides as obtained by (Ansari and Shrivastava, 1991; Purmima, *et al*., 1999 and Farid, *et al*., 2000), who reported that utilization of some plant originated essential oils gave better antifungal effect on soilborne pathogens than captan and benomyl.

Effect of biofertilizers on the development of damping-off disease infesting 3 cantaloupe hybrids:

Biofertilizers induced effectiveness on damping-off disease-infesting Cantaloupe (ranged from 66.6 in case of Rhizobacterin to 88.9% in case of phosphorin protection, while untreated control gave 44.4%) (Tables 5 and 6). When they compared with chemical fungicides, they gave lower effects than fungicides which gave100% protection. This result agree with (O'Gara et al., 1996, El Ghany 1996, Koreish et al., 1998, Mahmoud and Mahmoud, 1999) who found that some biofertilizers inhibited some soilborne fungal pathogens; *R. solani, A. alternata* and *F. solani.*

Effect of soil solarization in combination with methyl bromide on cantaloupe wilting disease caused by soilborne fungal pathogens under field conditions at Ismailia.

The results obtained reveal that the combination of soil solarization treatment for 10 weeks with the low dose 35 g methyl bromide / m^2 gave the same effect of methyl bromide at high dose of 70 g/m^2, it is a good step forward to reduce the chemical fumigation dose with methyl bromide in combination with soil solarization in controlling the causal organisms of cantaloupe wilt disease with 50% (Table 7), such results agree with (Bell et al., 1991), who found that soil solarization in combination with methyl bromide reduced seriously infection with weed seeds and soilborne pathogens including sudden wilt of melon caused by complex of fungi and also reduced tomato decline, they found promising results.

Table 5. Effect of mixing biofertilizers with soil on the disease incidence on cantaloupe hybrids caused by *F. oxysporum* f. sp. *melonis*

Ideal					Primal					Vicar					
Survival %	B		A		Survival %	B		A		Survival %	B		A		
	M%	N0	M%	N0		M%	N0	M%	N0		M%	N0	M%	N0	
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Non –infested
44.45	22.22	1.33	33.33	2.00	55.56	11.11	1.66	33.33	2.00	44.45	11.11	1.33	44.44	1.66	(Infested)
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	Thiram
100	0.00	3.00	0.00	3.00	0.00	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Topsin
55.56	22.22	1.66	22.22	2.33	66.67	11.11	2.00	22.22	2.33	55.56	11.11	1.66	33.33	2.00	*Phosphorin*
66.67	22.22	2.00	11.11	2.66	77.78	11.11	2.33	11.11	2.66	66.67	0.00	2.00	33.33	2.00	*Rhizopactrin*
77.78	11.11	2.33	11.11	2.66	66.67	22.22	2.00	11.11	2.66	77.78	0.00	2.33	22.22	2.33	*Mycrobin*
L.S.D 0.05 =				0.33					0.56					0.71	

Table 6. Effect of mixing biofertilizers with soil on the disease incidence on cantaloupe hybrids caused by *F. solani*

Ideal					Primal					Vicar					
Survival %	B		A		Survival %	B		A		Survival %	B		A		
	M%	N0	M%	N0		M%	N0	M%	N0		M%	N0	M%	N0	
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	Non –infested
44.45	22.22	1.33	33.33	2.00	55.56	11.11	1.66	33.33	2.00	22.23	33.33	0.66	44.44	1.66	(Infested)
88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	88.89	11.11	2.66	0.00	3.00	Thiram
100	0.00	3.00	0.00	3.00	100	0.00	3.00	0.00	3.00	0.00	0.00	3.00	0.00	3.00	Topsin
88.89	11.11	2.66	0.00	3.00	88.89	0.00	2.66	11.11	2.66	77.78	0.00	2.33	22.22	2.33	*Phosphorin*
77.78	0.00	2.33	22.22	2.33	77.78	22.22	2.33	0.00	3.00	55.56	11.11	1.66	33.33	2.00	*Rhizopactrin*
66.67	11.11	2.00	22.22	2.33	88.89	0.00	2.66	11.11	2.66	66.78	11.11	2.00	22.22	2.33	*Mycrobin*
L.S.D 0.05 =				N.S					N.S					N.S	

NO= Number of plants M % = Mortality percentage A= After 20 days from planting B = After 40 days from planting

Table 7. Effect of soil solarization compared with methyl bromide in different doses on cantaloupe sudden wilt

Treatments	Germination %	Wilted plant %
Methyl bromide 35 gm/m^2	91.3	4.86
Methyl bromide 35 gm/m^2 + soil solarization	92.7	3.79
Methyl bromide 70 gm/m^2	90.1	4.52
Soil solarization alone	91.1	17.70
Control	90	22.63
L.S.D 0.05 %	N.S	6.50

We hope that the obtained results will provide a starting point for discovering some non-chemical new alternatives of natural origins to avoid or reduce the consumption and harmfulness of chemical fungicides against soilborne fungal pathogens infesting Cantaloupe and other cucurbits.

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STRUCTURAL ANALYSES OF DISEASE PROGRESSION OF APPLE SCAB (*VENTURIA INAEQUALIS*)

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Apple scab, caused by *Venturia inaequalis* (Cooke) G. Wint., is the most prevalent disease in apple orchards in most apple growing areas. It causes damage to the leaf and fruit, which negatively affects yield and fruit quality. As environmental considerations have become increasingly significant, most European countries have changed their strategy in the selection of plant protection chemicals. In environmental-friendly horticulture, guidelines have been established, and control methods are applied in integrated and organic production systems (Anonymus, 1989; Dickler, 1992; Bloomers, 1994 and Miklay, 1995).

Many new methods, including microbial preparations, have been evaluated for efficacy against apple scab *in vitro* and *in vivo* (Burchill and Cook, 1970; Heye and Andrews 1984; Miedke and Kennel, 1990 and Benyagoub et al., 1998). Despite of these, little practical experience is available about the dynamics of disease progression in environmental-friendly production systems. Moreover, information is needed about cultivars under different ecological conditions and control methods, in order to evaluate the stability of host resistance.

The aim of our study was to analyse disease progression of apple scab on three apple cultivars in organic and integrated production systems.

Materials and Methods

Orchard site and disease assessment

The study was carried out in an experimental apple orchard at Debrecen-Pallag, Hungary, which was divided into two experimental blocks. One of the experimental blocks was treated according to the Hungarian IFP guidelines (Anonymous, 1995); and the other according to Hungarian organic production guidelines (Anonymous, 1997). These guidelines have been applied since 1997, when the orchard was planted. The experimental field consists of 40 apple cultivars. The cultivars were planted in randomised blocks with five replicates in the experimental field. Each block consisted of seven trees, but observations were only made on the middle five trees of each plot. Single trees were used as observation units.

The dwarf trees grafted on M26 rootstock were planted at a distance of 4 x 1.5 m and pruned to a spindle shape. Observations were made on cvs. 'Egri Piros', 'Royal Gala', and 'Elstar'.
Disease assessments were made on leaves and fruits in 2001 and 2002. For leaves, five sampling units were chosen at random, and for each unit, one-year-old lateral twigs were selected with 50 leaves. Each unit was tagged at the beginning of May, and the total number of healthy and diseased leaves was counted on each observation date. Twenty fruits were chosen at random for each observed tree at each observation date. Incidence of leaf and fruit were calculated. Assessments were made on a weekly basis from the beginning of May until mid-October.

Data analysis

Combined data of both years were used for the data analysis. Disease incidence data (dependent variable, corresponding to '*y*' axis) was linearised based on transformation functions of Hau and Kranz (1977). The transformation functions were: logarithmic (10 based): $z = \log(x)$, exponential: $z = \ln(x)$, Gompertz: $z = -\ln(-\ln(x))$, logistic: $z = \ln(x/(1-x))$, monomolecular $z = \ln(1/(1-(x))$. Time (independent variable, corresponding to '*x*' axis) was used without transformation. Linear regression analyses were performed for all linearised dependent variables against non-transformed independent variables. The best regression equations were selected by the following criteria:

- constants and coefficients with reasonably small standard error;
- P-value < 0.1;
- as high R^2 (coefficient of determination) as possible.

For explaining structure of disease progression, only one transformation function was selected, which generally gave the best result for above selection criteria. Obtained linear regression equations were used to quantify the disease growth rate (*k*). Growth rates were obtained from slopes of linear regressions over time (Berger, 1981).

"Area under the disease progress curve" (AUDPC) was also calculated from the data points based on Naragajan and Muralidharan (1995).

Results

Regression analysis

Generally, the best function was the logistic transformation. Values of intercept, slope, standard errors (SE), coefficient of determination (R^2) and mean standard errors (MSE) of obtained linear regression equations can be seen in Table 1. The R^2 showed generally high values (0.8), standard errors were reasonably small in the case of leaf incidence. Disease growth rate (*k*), obtained from the slope of linear regression equations, showed the highest values on leaf incidence of cvs. 'Gala Must' and 'Elstar' in the organic production system. Adequate disease growth rate of leaf incidence was higher in the organic production system than in the integrated one. Disease growth rates of fruit incidences were low and with negative signs on cvs. 'Elstar' and 'Egri Piros'.

Area under the disease progress curve

In all cases, AUDPC were higher in the organic than in the integrated fruit production system. AUDPC showed great differences in leaf incidences among cultivars (Table 2). Differences in leaf incidences were the largest in the organic production system. AUDPC values of leaf incidence for susceptible cultivars were twice to ten times higher compared to old or resistant cultivars. Great difference was found in the AUDPC values of fruit incidence in the organic production system. Cultivar differences in AUDPC values were smaller in the integrated production system.

Discussion

The present study analysed epidemic disease procession of apple scab on three apple cultivars in two environmental-friendly production systems.

Disease growth rate (*k*), obtained from slope values of linear regression equations, was variable depending on production system, cultivars and plant organs (Table 1). Analytis (1973) demonstrated that the rate of disease increase varied from 0.1 to 0.34 in the number and diameter of scab lesion on an individual leaf. Disease growth rate of leaf incidence ranged from 0.018 to 0.044 in this study. There is no scientific information about disease growth rate of apple scab under different fungicide treatments, but results of Plaut and Berger (1981), Gregory et al. (1981) and Rouse et al. (1981) supported our findings on other diseases. They found that if epidemics begun from lower levels of initial disease, then early disease progression was increasingly faster or the disease growth rate was increasingly higher. In this study, disease growth rate for leaf incidence provided permanent disease progression with a high correlation. In contrast,

Table 1. Linear regression analyses of disease progression of apple scab on three apple cultivars in integrated and organic apple production systems, Debrecen - Pallag, 2001-2002

System[a]	MSE[b]	R^{2} [c]	Intercept[d]	SE_i[e]	Slope[f]	SE_s[g]	F-test[h]
			Gala Must				
ORG fruit	0.013	25.1	-2.11	± 0.049	0.001	± 0.001	*
ORG leaf	0.111	90.5	-4.12	± 0.115	0.033	± 0.002	***
INT leaf	0.306	78.5	-4.22	± 0.766	0.014	± 0.003	**
			Elstar				
ORG fruit	0.032	62.4	-1.26	± 0.092	-0.004	± 0.001	*
ORG leaf	0.114	91.4	-3.69	± 0.222	0.025	± 0.004	**
INT leaf	0.111	82.7	-5.13	± 0.673	0.021	± 0.007	**
			Egri Piros				
ORG fruit	0.027	32.5	-1.59	± 0.045	-0.003	± 0.001	*
ORG leaf	0.114	91.3	-3.23	± 0.113	0.019	± 0.003	***
INT leaf	0.142	81.3	-3.85	± 0.387	0.009	± 0.001	**

[a] ORG fruit = fruit incidence data in the organic production system, ORG leaf = leaf incidence data in the organic production system, INT fruit = fruit incidence data in the integrated production system, INT leaf = leaf incidence data in the integrated production system.
[b] MSE = mean standard error.
[c] R2 = coefficient of determination.
[d] Slope value is the coefficient of linear regression analysis and the disease growth rate (*k*) of disease progress.
[e] SEi = standard error of intercept.
[f] Intercept is the constant of linear regression analysis.
[g] SEs = standard error of slope.
[h] F-test = *** < 0.01, ** 0.01 - 0.05, * 0.05 - 0.1, ns > 0.1.

Table 2. Area under the disease progress curve (AUDPC) of apple scab on three apple cultivars in organic and integrated fruit production systems, Debrecen - Pallag, 2001-2002

Cultivars	Integrated		Organic	
	leaf	fruit	leaf	fruit
Gala Must	865.2 a[a]	0	2,811 a	1, 345 a
Elstar	327.4 b	0	2,134 a	879.2 b
Egri Piros	243.7 b	0	1,089 b	103.5 c
F-test[b]	***	-[d]	***	***
SED (df =24)[c]	42.22	-	402.4	121.4
$LSD_{0.05}$	89.23	-	821.3	261.5

[a] Values within columns followed by different letters are significantly different.
[b] F-test = *** < 0.01, ** 0.01 - 0.05, * 0.05 - 0.1, ns > 0.1.
[c] SED = standard errors of differences of mean values, df = degrees of freedom.
[d] Because of zero values, no F-test, SED and LSD values were available.

disease growth rate for fruit incidence support a slow disease increase or, in some cases, slow disease decrease in fruit scab epidemic progression. The reason for this is that some of the early-infested fruits had fallen and the ontogenic resistance of fruits steadily increased during the growing season. Consequently, the percentage of diseased fruits was somewhat lower through the summer and in early autumn, compared to the spring disease level.

Although the disease growth rates were quite similar for both leaf and fruit in both production systems, the “area under the disease progress curve” showed great differences in both production systems. Plank (1963) and Kranz (1974) found close correlation between the disease growth rate and the AUDPC for several diseases. In this study, AUDPC referred to the effectiveness of fungicides and level of epidemic in both production systems. Moreover, AUDPC gave more differences for comparison of disease progression than disease growth rate in both integrated and organic production systems.

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Summary

STRUCTURAL ANALYSES OF DISEASE PROGRESSION OF APPLE SCAB (*VENTURIA INAEQUALIS*)

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Disease progression of apple scab was analysed in a two-year-study, in two environmental-friendly production systems (organic and integrated) on cvs. ‘Gala Must’, ‘Elstar’ and ‘Egri Piros’. Linear regression analysis of transformed disease incidence data and “area under the disease progress curves” (AUDPC) were used to analyse the epidemic processes on the three apple cultivars. In linear regression analysis, the best function was the logistic transformation. Disease growth rates of leaf incidence were higher in the organic production system than in the integrated one. Disease growth rate of fruit incidences was low and with negative signs on cvs. ‘Elstar’ and ‘Egri Piros’. AUDPC showed great differences in leaf incidences among cultivars and between production systems. AUDPC gave more possibility for comparison of disease progression than disease growth rate in both integrated and organic production systems. Results were compared with similar studies on different pathosystems and biological interpretations of the analyses are discussed.

PHYTOPHTHORA ROOT AND CROWN ROT OF FRUIT TREES IN BULGARIA

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In the literature diseases caused by *Phytophthora* species are known as "Phytophthora root and crown rot" (Ellis, 1997; Teviotdale and Gubler, 1999); "crown collar rot" (Hickey and Yoder, 2001); "Phytophthora root, crown and collar rots" (Wilcox, 1998). They are spread worldwide and have broad range of host plants – fruit trees, citrus, forest and park species. Recently *Phytophthora*s cause serious damages on apples, cherries, apricots, peaches (Jeffers and Aldwinckle, 1987; Ellis, 1997; Growe, 1997; Teviotdale and Gubler, 1999; Wilcox, 1990, 1998). Pear and plum trees appear relatively resistant.

Tree decline and dying caused by *Phytophthora* root and crown rots are frequently misdiagnosed as suffering from "wet feet" (root asphyxiation) and sometimes confused with those suffering from winter injury (Ellis, 1997).

Symptoms expression depends upon how much of the root or crown tissues are affected, and how quickly they are destroyed. Crown rots advance rapidly and trees collapse after the first warm weather period in the spring (Teviotdale and Gubler, 1999). Leaves wilt, dry and remain attached to the tree. *Phytophthora* infections typically kill young trees.

Ellis (1997) states that wet soils that remain saturated for extended periods are required for disease development. Above-ground symptoms vary between tree species but include generally reduced tree vigor and growth, yellowing or chlorosis of leaves, and eventually collapse or death of the tree. Infected trees may decline slowly over one or more years, or they may collapse in spring. Trees may also appear healthy in spring but die suddenly in the latter part of the growing season. Below-ground symptoms include reddish-brown discoloration of the inner bark and wood. A sharp line demarcates the diseased and healthy portion of the crown. Similar symptoms can be found on the roots (Ellis, 1997; Hickey and Yoder, 2001).

Periods of 24 hrs or more saturated soil favourable for *Phytophthora* infections. Conversely, good soil drainage and more frequent but shorter irrigations reduce the risk of root and crown rot (Teviotdale and Gubler, 1999). Disease is more often observed in low areas of the orchard on heavy, poorly drained soils / Hickey, Yoder, 2001; Wilcox, 1998/.

Phytophthora root and crown rots are caused by several *Phytophthora* species. All of them require extremely wet soils in order to infect and cause significant damage but they differ in destructiveness depending on plant

species (Ellis, 1997). Rootstocks vary in susceptibility to different *Phytophthora* species. Among apple rootstocks, seedlings are relatively resistant. Among dwarfing-apple rootstocks M-9, M-2 and M-4 are relatively resistant. The Canadian rootstock Ottawa-3 has M-9 type resistance. M-7 and MM-111 are moderately susceptible, M-26 and MM-106 are susceptible, MM-104 is highly susceptible (Ellis, 1997; Wilcox, 1998). Teviotdale and Gubler (1999) wrote that MM-104 and MM-106 are more susceptible than M-9 and M-26. M-111 is susceptible to moderately resistant, M-7 a susceptible (Yoder and Biggs, personal observations). Among stone fruits, plums are relatively resistant. Mahaleb is the most susceptible cherry rootstock, whereas Mazzard, Morrello, and Colt are more resistant and recommended on heavier soils (Ellis, 1997). Fungus infection is favoured also by cool soils (10-16 ^{o}C) (Hickey and Yoder, 2001) for some *Phytophthora* species. For other soil temperature in the range of 15 to 25^{o}C is appropriate (Wilcox, 1998).

Phytophthora fungi in fruit trees are difficult to control. Teviotdale and Gubler (1999) recommend foliar application of Aliette (fosetil-Al) and soil application of Ridomil Gold (mefenoxam) in early spring and fall.

Soil fumigation is considered ineffective because it never completely eradicates the fungus, and *Phytophthora*s are easily reintroduced into soils (Ellis, 1997; Wilcox, 1998). Fungicides can be effective when used preventively but not in infected trees showing moderate symptoms. Wilcox (1998) also recommends fungicide treatments in the early stages of decline, in advanced stages is cheaper to remove the tree and replant.

In general fungicides are most effective when used in combination with cultural practices, including comparatively resistant rootstock and drained soils.

Materials and Methods

In the period 1999-2002 plant materials from the following regions have been analyzed:

- Bjaga /Peschera, Plovdiv/ - apple trees
- Katunitza /Plovdiv/ - cherry rootstocks
- Trilistnik /Plovdiv/ - apple and cherry trees
- Brestnik /Plovdiv/ - cherry trees

The causal agents have been isolated on specific culture media and applying “baiting bioassay” method. Culture media are based on corn meal agar plus pimaricin, ampicillin, rifampicin, PCNB and hymexazol (PARP). Both methods are standard ones for isolating *Phytophthora* fungi from infected wood and soils. Wood showing symptoms (from the root or crown zones) have been thoroughly washed with running water. Afterwards it has

been surface sterilized with alcohol, and cut into small pieces. They have been plated into Petri dishes on PARP media.
Sterilized woody tissues have been used in baiting bioassays also. Green apples serve as "trap culture". They have been surface sterilized, afterwards sterile cone shape cuts have been done. In holes on the fruit diseased pieces from wood have been placed, 4-5 ml sterile water have been added, wax and parafilm as a cover. Infected fruits have been incubated in growth chamber (25^{o}C, RH 75% and 12 hrs photoperiod) for 7-10 days, till symptoms appeared. Fruit tissues from the edge of rotten zone have been transferred on V-8 or PDA media.
PDA has been used for studying morphological and cultural characteristics of mycelia and conidia, as well as formation of chlamydospores, antheridia, oogonia and oospores.
Strains isolated from apple trees (Bjaga, Plovdiv) and cherry rootstocks (Katunitza, Plovdiv) have been identified based on morphology and studied on culture media (PDA, V-8).

Results and Discussion

In Bulgaria first symptoms have been found during 1998-99 on 2-3 years old apple trees in the region of Peschera (Bjaga) and on 2-year-old cherry rootstocks (Katunitza, Plovdiv). Latter in the period 2000-2002 the same type of symptoms, caused by *Phytophthora*, have been observed on fruit trees in two more locations close to Plovdiv – Trilistnik (2-year-old cherries) and Brestnik (cherries).
Symptoms of Phytophthora root rot type disease appear in early spring. Infected trees suffer bud break delay, their leaves are small and chlorotic, branches dye all of a sudden or whole tree dry out. That type of symptoms are not enough for diagnosis, but point out that root system or crown of the tree can be affected. Reddish-brown lesions with wet, necrotic appearance have been found on the crown and roots of infected trees (Fig 1). There is distinct margin between diseased and healthy wood tissues. Latter infected wood becomes dark brown. Lesions can spread as a ring and also upwards on the trunk of the tree.
Diagnostic symptoms are found also bellow-grafting zone as large dark spots clearly defined from healthy tissues. When bark is removed orange to reddish discoloration has been seen and a dark line marks the border with healthy tissues. Roots show similar symptoms when infected.
Trees with healthy appearance in spring can all of a sudden wilt and dry out later in the season (mainly in August-September). When trees are infected leaves drop down early and have reddish discoloration at the end of August.
Studies point out that disease spread is favoured by wet and heavy not well-drained soils. Heavy rains also provoke disease symptoms.

Causal agents have been isolated from infected wood on PARP media and applying “baiting bioassays”. Identification has been done on PDA based on fungal morphology and cultural characteristics.
“Apple” strains isolated from apple trees differ in appearance. First group on PDA develops whitish, slightly smoky, featherlike colonies with radial growth (star-like) (Fig. 2). Sporangiophores are simple or branched, zoosporangia are oval, fusiform or irregular in shape (38-40 x 31-33 μm^2).

Figure 1. Symptoms of Phytophthora root and crown rot on apple trees (Bjaga region)

Terminal chlamydospores are rarely formed, especially when NH_4NO_3 has been added to media. Oospores have been developed. Second group has whitish, smoky, fluffy mycelia (Fig. 3).

Figure 2. Colony of *Phytophthora citrophthora* isolated from apples (Bjaga region)

Figure 3. Colony of *Phytophthora cactorum* isolated from cherries (region Katunitza)

Mycelia are long, branched, normal or slightly swollen at the point of branching. Conidiophores are simple or sympodially branched. Zoosporangia are oval to elongated (lemon shaped), average size 33-34 x 26-28 μm on a media. After long period of culturing (more than 7 days) terminally or intercalary chlamydospores developed.
Antheridia and oogonia, as well oospores are formed in large numbers on strains with fluffy mycelia.They are sparse in strains with radial growth of the colonies.
"Cherry strains" isolated from cherry trees have whitish, slightly smoky colonies in appearance, with fluffy aerial mycelia. On mycelia oval or lemon shaped (elongated) zoosporangia (conidia) are formed, as well as abundant chlamydospores and oospores.
Data analysis and their comparison with results published by other authors (Smith, 1937, 1953, 1955, 1956; Novotelnova, 1974; Ellis, 1997; Hickey and Yoder, 2001; Smith and Smith, 1906, 1925, etc.) let us to conclude that strains isolated from apples belong to the species:
Phytophthora cactorum (Leb. & Cohn) Schröt and
Phytophthora citrophthora (R.E. Smith & E.H. Smith) Leonian.
Pathogen isolated from cherries belongs to *Phytophthora cactorum* (Leb. & Cohn) Schröt.

Conclusions

Analyses of results received from the studies on morphological and cultural characteristics of the strains isolated give us evidence to conclude that two *Phytophthora* species have been found on fruit trees in Bulgaria: *Phytophthora cactorum* (Leb. & Cohn) Schröt – on apples and cherries and *Phytophthora citrophthora* (R.E. Smith & E.H. Smith) Leonian – on apples.

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Summary

PHYTOPHTHORA ROOT AND CROWN ROT OF FRUIT TREES IN BULGARIA

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In Bulgaria disease first have been recognized during the autumn of 1998-1999 on young apple trees in Plovdiv region (Bjaga, Peschera) and on 2-year-old cherry root-stocks (Plovdiv, Katunitza). Later in the period 2000-2002 symptoms have been found on 2-year-old cherries (Trilistnik, Plovdiv), cherry trees (Brestnik, Plovdiv) and on young apples (Kjustendil).
In the literature diseases caused by *Phytophthora* fungi are known as Phytophthora root and crown rot; crown collar rots; Phytophthora root, crown and collar rots respectively. *Phytophthora* species are world-wide spread on large number of host plants. Recently they cause considerable damages on apples, cherries, apricots and peaches.
Disease symptoms have been studied with the aim of diagnostic and in connection with conditions favouring its spread and appearance.
Phytophthora species have been isolated on specific artificial media containing antibiotics (PARP) and also by applying "baiting bioassays" on green apples.
Cultures isolated from apples have been identified as *Phytophthora cactorum* and *Phytophthora citrophthora*. From cherries only *Phytophthora cactorum* have been isolated.
Morphological and cultural characteristics (morphology of mycelia, conidia, their shape and size, presence of chlamydospores, antheridia, oogonia, oospores) have been studied on different nutrient media.

INCIDENCE OF *THIELAVIOPSIS POPULI* ON HYBRID POPLARS IN HUNGARY

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Unusual root disease of *Populus* x *euramericana* "Pannónia" was observed in summer 2002 in Hungary, near Bábolna. Beside the *Fusarium* species, *Thielaviopsis populi* (Veldeman ex Kiffer et Delon) Paulin, Harrington et McNew was isolated from the roots and collar of decaying trees. This is the first record of *T. populi* in Hungary.

Thielaviopsis populi (=*Chalara populi* Veldeman ex Kiffer et Delon) is a conidial fungus pathogen on poplars. It was described first in Belgium as causing bark lesions killing the cambium of the stem (Veldeman, 1971). The fungus shows some similarities with the conidial state of *Ceratocystis fimbriata* Ellis and Halsted, ubiquitous ascomycete reported also from *Populus tremuloides* in North America and from poplar hybrids in Poland (Gremmen and Kam, 1977). Nevertheless some differences were detected both in the morphology (Gremmen and Kam 1977) and in the ITS sequences of the nuclear rDNA (Paulin-Mahady et al., 2002) of the two fungus.

The field investigations were effected in August and September 2002 in two 3- and 4-year-old stands, respectively. The observed symptoms were: swollen and cracked collar usually wounded by xylophagous insects, the bark of the roots turned dark coloured, soft and rot, then the attacked trees died.

Pathological materials and soil samples were collected from the collar and roots of decaying trees in different state of decay. The material was washed, superficially disinfected with chlorine, cut into pieces and incubated in wet chamber. After a few days the grown fungal colonies were examined and identified. The isolations were performed on PDA media with plant tissues from the limit of necrosis and from the colonies developing in wet chamber. Simultaneously special isolation essays were made from soil and roots for detection of *Phytophthora* species, their presence being presupposed on the ground of similar disease symptoms.

Mostly different *Fusarium* species were identified on the samples with advanced symptoms. Bacteria and nematodes were also abundantly detected. On the wood and bark pieces with initial symptoms *Thielaviopsis populi* was identified and isolated. On the wood, perithecia of *Ceratocystis*

type developed in this case but in culture the formation of perithecia was not observed. The presence of *Phytophthora* was not proved.

The morphological characters of *T. populi* isolate (culture, endoconidia and conidiophores as well as chlamydospores) were identical to the descriptions of the species in the literature cited. The molecular identification was performed by T. C. Harrington. Our isolate proved to be identical in ITS sequences of the nuclear rDNA to other isolates of *T. populi* in his collection.

The pathogenicity probe was conducted by inoculation under the bark of hybrid poplar cuttings. In average 25.12 x 12.75 mm large necroses were produced in the bark at 5 weeks after inoculation. For comparison, under the same conditions the *Fusarium* isolates caused far larger necrosis extending to the full surface of the cuttings of 45 cm. This fact indicates that *Fusarium* species had an important role in the decay of the trees. *T. populi* could be isolated more rarely and from the initial symptoms only, and produced smaller necrosis by artificial inoculation.

Regarding the ethiology of the decay the presence and mass attack of the xylophagous insects as poplar hornet clearwing, (*Aegeria apiformis*), large poplar longhorn (*Anaerea carcharias*) and poplar borer (*Agrilus populneus*) at the collar of trees should be considered as inciting factor, which opened infection portals for the pathogens (*Fusarium* species, *T. populi*) causing bark necrosis of the root system.

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Summary

INCIDENCE OF *THIELAVIOPSIS POPULI* ON HYBRID POPLAR IN HUNGARY

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Unusual root disease of *Populus x euramericana* "Pannónia" was observed in summer 2002. *Thielaviopsis populi* was isolated from the roots and collar of decaying trees. The artificial inoculations proved the pathogenicity of the isolate. This is the first record of *T. populi* in Hungary.

TISSUE DEFORMATIONS OF SUNSCALD INJURY ON THE SURFACE OF APPLE FRUIT (*MALUS DOMESTICA* BORKH.) AND ITS METEOROLOGICAL CAUSES

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Sunburn of apple fruit is a surface injury caused by solar radiation, that in the initial phase results in a light corky layer, golden or bronze discoloration and injuries of the epidermal tissue on the surface exposed to radiation. According to the definition of Retig and Kedar (1967) "sunscald" is the physiological injury of the fruit that significantly affects its quality. Meteorological elements, the physiological condition of the plant, its variety may all contribute to the formation of the injury. The change mainly occurs in the surface layers or those close to the surface. Later, plant pathogens such as *Alternaria tenuis, Physalospora obstusa, Monilia fruticola, Monilia laxa, Monilia fructigena, Glomerella cingulata, Venturia inaequalis* can infect the apple fruit through the injured epidermal tissue making the fruit unmarketable (Gurnsey and Lawes, 1999; Holb, 2002; Leeuwen *et al.* 2000, 2002). Therefore, this phenomenon causes serious economic losses in apple plantations (Brooks and Fisher, 1926; Ware, 1932; Meyer, 1932; Whittaker and McDonald, 1941; Moore and Rogers, 1942; Barber and Sharpe, 1971; Bergh et al., 1980; Simpson et al., 1988; Warner, 1997; Schrader et al., 2001).

Barber and Sharpe (1971) studied the injuries on pepper and pumpkin fruits and separated three sunscald types: heat injury sunscald -HIS, ultra-violet radiation sunscald -UVS and photo-dynamic sunscald of heated tissues -PSHT. Besides the above-mentioned grouping, American apple growers divide the phenomenon called sunscald into three groups. They distinguish "sunburn", "sunscald" and "delayed sunscald". Previously, Walker (1952, 1957) used the phrase of "sunscald" for all injuries caused by solar radiation. On the basis of the triggering reasons Schrader et al. (2001) determined two main types of "sunburn" injuries. The so-called "sunburn necrosis" is caused by the effect of heat and necrosis of the epidermal and sub-epidermal cells. This phenomenon causes spottiness on the sunlit side of the apple fruit. The second type is called "sunburn browning" and causes yellowish or brownish patches on the sun exposed side of the apple fruit. Schrader *et al*. (2001) also determined the physiological reasons of the two phenomena. Sunburn necrosis already happens when the overheating of the surface of the fruit reaches 52±1ºC, meanwhile the permeability of cell

membranes is damaged. Sunburn browning forms at 46-49ºC surface temperature, but sunlight has a decisive role in its formation as well. In this case the membranes of the surface cells of the apple fruit are injured in a lesser extent (Schrader et al., 2001, 2003).
The phrases of "sunburn" and "sunscald" are often mixed in the common knowledge.
The American Phytopathological Society defines sunburn as a fruit injury caused by solar radiation, while sunscald as injuries occurring on the surface or in sub-surface layers caused by freeze (Jones and Aldwinckle, 1990).
In Hungary and Slovakia, Vanek and Szőke (1988) detected serious sunburn on apple and grape fruits caused by solar radiation. They predicted an increasing sunburn damage for the following years because of increasing UV-light radiation and global warming. Due to the injury, the amount and quality of yield significantly decrease many times. According to Arndt (1992) in the case of the Jonagold variety this can cause a 50% loss in yield, but this variety is sensitive to sunburn. Even in the beginning of ripening on the surface of the fruit discoloration caused by solar radiation, i.e. surface scars may appear, that have influence on the further coloration as well as taste, then marketability and storability of the fruit. Gurnsey and Lawes (1999) determine that under American market conditions the excellently coloured apple is worth even 3-4 dollars more per hamper. Schrader et al. (2001) report on several-million-dollar loss in the apple plantations in America.
More detailed knowledge of the process of sunburn can contribute to the estimation of the risks caused by climatic factors, even to each variety. The obtained information can help to decrease the further losses, when planning fruit plantations (e.g. direction and distance of rows, irrigation, shaping of the structure of canopy). With proper agri-technical methods the frequency of the occurrence of sunburn can be decreased in apple plantations (Meheriuk et al., 1994).

Description of symptoms of injuries caused by solar radiation

"Sunburn" causes golden-bronze discoloration on the sunlit side of the apple fruit. Thus, it detracts from its appearance, but in most of the cases it would not cause serious damages in the epidermal tissue. Even the sub-epidermal tissues show no serious change. The sunlit area of the fruit is firmer, but it tends to soften quickly during storage (Gurnsey and Lawes,1999). True "sunscald" occurs when the fruit growing in shade is suddenly exposed to strong sunlight. Due to sunlight, light or yellowish-brown patches appear on the apple fruit's surface and in its appearance more serious damages can happen in the surface tissues than in the other case. This damage is most common on fruits on the southern, west-southern sides of the tree. These

symptoms can be observed on apple fruits fallen from the tree and those ones in hampers as well, if they were exposed to strong radiation for long periods. During storage or sometimes even already on the tree brown, hard, sagging patches with bright surface appear, which have spongy structure inside. These are called "delayed sunscalds", that mean entrance points for fungi (e.g. Alternaria rot) (Barber and Sharpe, 1971; Bergh et al., 1980; Simpson et al., 1988).
The more severe form of this damage indicates serious changes in the cuticle, in the epidermal and the sub-epidermal tissues. The cell walls thicken. The volume of phenols increases in the intercellular space and the structure of plastids and thylakoids alters (Barber and Scharpe, 1971; Andrews and Johnson, 1996, 1997).

Reasons of sunburn, conditions of its formation

Besides the basic role of solar radiation, other factors play a role as well in the formation of sunburn. According to Barber and Sharpe, (1970) basically the following factors influence the formation of sunscald: solar absorptivity, interception of solar energy, temperature tolerance, "specific photostability", tolerance to ultra-violet radiation and degree of adaptation or sensitisation to the environment. It occurs mainly in such areas where the air temperature is high and during the ripening period the number of sunny hours is high as well. These damages also occur in great number when cool or mild weather situations are followed through a short transition by a hot, sunny period. If the change does not occur immediately, the plant can acclimatise to the changed climatic conditions, therefore, the risk of sunscald decreases as well. The damage is intensive, if it is accompanied by water stress as well in this period (Brooks and Fisher, 1926; Ware, 1932; Meyer, 1932; Whittaker and McDonald, 1941; Moore and Rogers, 1942; Barber and Sharpe, 1971).

Biotic reasons of sunscald formation

The main key factors during the formation of sunscald are primarily the variety of the apple, its physiological state and the structure of the planting.
Apple varieties are sensitive to solar radiation and temperature to a different extent. This originates from the differences in environmental needs, but the tissue structure of the fruit, thickness of cuticle and wax as well as the pigmentation characteristic to the variety has important roles too. During each stage of ripening, sensitivity to solar radiation and temperature can change. This can be explained by the tissue development of the flesh of the fruit.
Some varieties – for example the Granny Smith – are sensitive to light since their epidermal tissue is thin, therefore, this can be damaged more easily.

The lack of calcium (soils with calcium deficiency) increases sensitivity to light since it has an effect to the thickness of the epidermal tissue. Under physiological state of the plant, we mean water- and nutrient supplies. Transpiration heat loss can decrease the overheating of the fruit, the effectiveness of which lessens in dry periods (draught) or when there is small accessible water supply (sandy soils). Then, the risk of sunscald increases. Due to the effect of the not properly distributed nutrients, the plant tissues become more sensitive to injuries. Nitrogen stimulates the production of new shoots that is disadvantageous for reaching the optimal colour because of shadowing (Meheriuk et al., 1994).
Brooks and Fisher (1926) and Meyer (1932) established that apple varieties having red-colour fruits are more resistant to sunscald.
For formation of the intensive apple colours 20-25ºC daytime- and about 18ºC evening/night temperature are needed in the pre-harvest weeks. For formation of the proper colour 50-70% of the light that reaches the soil surface (full sunlight) is needed (Gurnsey and Lawes, 1999). In order to achieve this, the canopy of the tree is trained, a part of the new shoots is pruned during summertime (e.g. the variety of Royal Gala needs this pruning). The aim is to prevent the leaves from shading the ripening fruits. But the plantings and tree structure optimal for colour formation can increase the risk of sunscald (Gurnsey and Lawes, 1999). One of the protective mechanisms of the plant against light is that on the sunlit side the amount of the pigments (flavonoids, carotinoids, anthocyans) increases in the fruit skin. This process means a natural protection against solar radiation. These materials are also responsible for the taste and pattern of the apple fruit (Reay and Lancaster, 2001; Merzlyak et al., 2002).

Abiotic reasons of sunscald formation

According to the establishment of Smart and Sinclair (1976) in the case of grape the formation of sunscald depends on the direction of the wind, the velocity of the wind and the intensity of turbulence. The injury can particularly occur when the sunlit side of the fruit overheats (particularly in the case of fruits on the southern or south-western quadrant of the tree) and due to tissue injury, scars develop. From the environmental parameters, radiation flux density and the velocity of the wind can primarily determine the temperature of the fruit, but the size of the fruit, its albedo, the transpiration of the fruit and the heat change by long-wave rays play a role as well. This connection is based on the energy formula of the surface and estimates the maximum and minimum heat increase of the surface of the fruit exposed to sunlight. The variables that form the connection are the following: the absorbed radiation flux density, the size of the fruit, its heat conduction and the convective heat change coefficient (Smart and Sinclair,

1976). This latter can be calculated from the velocity of the wind on the basis of the formula of Nobel (1975).
The formation of sunscald can be determined by the weather conditions of several days, if the changes occur in a sensitive stage of the development of the fruit. Besides the elements of solar radiation, the other important change is that the temperature of the surface of the sunlit side of the fruit can be as much as 18ºC above air temperature and 8-9ºC warmer than that of the shaded side (Meheriuk et al., 1994). When the cool night is followed by a too hot daytime the anthocyanin synthesis strongly decreases. According to Arndt (1992), if the temperature in July, August and September surpasses the 28-32ºC, the formation of sunscald is more frequent as well. Barber and Sharpe (1971) and Schroeder (1961) studied the further effects of air temperature in the case of different fruits. Brooks and Fisher (1926) reported that if the surface temperature of an apple exposed to sunlight is as mush as 14ºC above the air temperature, the injury already appears. It occurs due to heat, not because of solar radiation. On the contrary Rabinowitch et al. (1974) established that in the case of tomato the phenomenon of sunburn occurs due to heat and visible light.
On the surface and in the flesh of the fruit the uneven distribution of light and temperature triggers a series of biochemical processes, while it changes the water management of the juicy fruit as well.
Smart and Sinclair (1976) studied the above-mentioned statements in the case of grape. Determining the energy of the absorbed solar radiation the following values were taken into consideration: convective energy loss, net energy loss because of long-wave radiation, energy loss of transpirational cooling and the energy loss led into the fruit. This was applied to a fruit having a small surface (grape). In the case of the applied connection two limitations have been mentioned. On the one hand, their model supposes homogeneous conditions in the fruit's flesh that cannot be considered as a mistake in the case of a fruit having high heat conductivity (e.g. grape). However, in the case of fruits with bigger diameter or having small heat conductivity this can be a problem. The other disputable part of the model used by Smart and Sinclair (1976) is that it takes the heat transformation coefficient uniform above the fruit surface (Thorpe, 1974).
Schrader et al. (2001) established in their trials conducted between 1996-1997, that UV-B radiation is not required for sunburn and cannot cause sunburn-like symptoms alone.

Decreasing of occurrence of sunburn by agri-technical methods

Applying these methods we should strive to decrease the exposure of fruits to heat and radiation, meanwhile the development of intensive colours characteristic to the species would not be hindered as well.

Summer pruning must be performed in such a way that the fruits' extensive exposure to radiation is avoided. Besides, overhead sprinkling can be applied in the canopy level to cool the surface of the fruits, but this can increase the possibility of spreading other infections (e.g. apple scab and fire blight).

In cool-houses during storage the development of delayed sunscald cannot be influenced. According to the studies washing with diphenylamine did not help much. In this case, the regular sorting can be a solution, since applying this we can decrease the possible infection spots of *Alternaria* rot (Meheriuk et al., 1994).

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PROPOSAL FOR THE EXPLOITATION OF RESISTANCE GENES IN PROTECTION OF THE HUNGARIAN WHEAT CULTIVARS AGAINST RUSTS

(Summary)

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Leaf rust *(Puccinia triticina)*, yellow rust *(P. striiformis)* and stem rust *(P. graminis)* are important pathogens in wheat growing areas in Hungary. The best method of the protection of wheat against rusts is to increase the resistance of cultivars.

The role of resistance genes in protection of wheat against rusts depends on virulence changes of pathogens. According to our data of seedling and field tests conducted in the past years (2000-2002) a lot of resistance genes (e.g. Sr31, Sr36, Lr9, Lr19, Lr24, Lr28, Lr29, Yr18) were effective against wheat rusts, which could give protection in wheat specifically in Hungary. Although our experiences proved that Hungarian cultivars carry only some of the effective resistance genes. This is the reason why nowadays rust epidemic can occur (e.g. yellow rust epidemic in 2000) in our country.

We have to increase the opportunity of protection of wheat against rusts by means of resistance genes in the future.

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EFFECT OF N-PHENIL-PHTALANIC ACID (NEVIROL 60 WP) ON QUANTITATIVE AND QUALITATIVE PARAMETERS OF SOME HORTICULTURAL PLANTS

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Ensuring yield-balance - although the applied technologies give a good possibility for this - in the large-scale farming is a difficult and complicated task. Pollination of certain horticultural species - because of climatic or genetic influences - is not possible in many cases. For sufficient yield amount and required yield quality we have to interfere in pollination. With the help of the N-phenyl-phthalanic acid, which is an agent of NEVIROL 60 WP, we can achieve this goal (Búza, 1986).

The N-phenyl-phthalanic acid as a regulator increases the working life of stigma, helps the better pollination, which results in a higher yield. The acid is not auxin, but it has synergistic effect with auxin in biological tests (Nyéki, 1980). Applying NEVIROL 60 WP, the possible unfavourable effects of the objective (agronomics, agrotechnics, species, weather, etc.) and subjective conditions of production can be reduced, yield fluctuation can be levelled, thus crop safety can be considerably increased. The product, like other regulators and all synthetic pesticides, is not approved in the organic production system (Holb and Heijne, 2001; Holb *et al*., 2003).

Its application is recommended at the flowering period in greenhouses, foilhouses as well as in field cultivation for some crops (tomato, paprika, peas, beans, cucumber, grape, apple, sour cherry, lupin, soya etc.) (Eõri, 1984; Teleky, 1985; Teleky and Bésán, 1986; Teleky and Eőri, 1984; Teleky and Horváth, 1986; Teleky and Veress, 1986). Its spray application at full bloom does not influence each crop parameter such as taste, colour, germinative ability, oil content etc. (Nyéki, 1980). The formulation with recommended rate irrespectively of culture and application conditions has not caused any phytotoxicity or partenocarpia. The product may be mixed with insecticides, fungicides and foliar fertilizers except for alkaline products. Attention has to be paid to phytotoxic effect of some scab fungicides at full bloom period, which should be avoided (Holb, 2002). The preparation of the spray liquid does not need any special measures as the preparation contains the necessary constituents to ensure quick and thorough

wetting. At applications with ground machine use 400-1000 l/ha, at aerial applications 60-80 l/ha.
It is important to note that while a higher yield is aimed at through the better fruit setting, a higher level of basic nutrition should be provided (Szirtes, 1984).

Materials and Methods

Field conditions of research and different characteristics of plant species are presented in Table 1. and Table 2. We have chosen four plant species, two of them were fruits and the other two were vegetables. The fruit species were apple and grape and the vegetables were tomato and cucumber. From each species we have chosen two varieties, one was fertilized and the other was without fertilization. The conditions of fertilization can be seen in Table 3.

Table 1. Field conditions of research

Genus	Variety	Place of experiment	Size of area (ha)	Size of parcel (m^2)	In-row spacing (cm)	Harvest	
						first date	last date
Apple	Idared	Nagylapos	9,5	400	300*100	29. 09.	
	Jonathán	Nagylapos	4,5	400	300*100	20. 09.	
Grape	Muscat Ott.	Mád	2,7	220	180*60	21. 09.	
	Olasz rizling	Mád	8,0	220	180*60	09. 10	
Tomato	Robot	Kálmánháza	1,2	18	60*30	16. 08.	21. 09.
	Delta	Kálmánháza	3,4	18	60*30	14. 08.	21. 09.
Cucumber	Barbara F1	Nagylapos	1,0	20	200*20	11. 07.	02. 09.
	Profi F1	Nagylapos	1,5	20	200*20	11. 07.	02. 09.

Table 2. Experimental conditions of NEVIROL 60 WP

Date of treatments	Treatments (x times)	Treatments (% of flowering)	Mode of delivery
04. 30.	1	50%	Kertitox NA-10
26. 04.	1	55%	Kertitox NA-10
04. 06.	1	10%	Novatur 1507
09. 06.	1	10%	Novatur 1507
24. 07. and 09., 20. 08.	3	60-90%	Novor 1005
24. 07. and 09., 20. 08.	3	60-90%	Novor 1005
03., 19. 07. and 11. 08.	3	60-90%	Arumic
02., 17. 07. and 10. 08.	3	60-90%	Arumic

Table 3. Conditions of fertilization in research

Genus	Varieties	Fertilization			Date of fertilization	
		N	P_2O_5	K_2O	base	head
Apple	Idared/MM 106	72	25	25	27 January	2 May
	Jonathán/MM106	72	25	25	26 January	2 May
Grape	Muscat Ottonel	112	18	52	21 December	22 March
	Olasz rizling	112	18	52	21 December	22 March
Tomato	Robot	159	58	58	12 January	15 August
	Delta	159	58	58	12 January	17 August
Cucumber	Barbara F1	12	4	12	15 January	*every week
	Profi F1	12	4	12	15 January	*every week

*In the growing period (from 2 June to 1 September)

Results and Discussion

The detailed results of NEVIROL application can be seen in Tables 4-7. The research results can prove, that there is quite a big difference between the pollination rates of different varieties. These differences can be increased by N-phenyl-phthalanic acid, which is the agent of NEVIROL 60 WP.

Apple

The research results show that the pollination rate increased by 22.9% without any fertilization, due to the effect of fertilization, the pollination rate can reach 28.8%. As a result of the usage of NEVIROL 60 WP, both the mass and diameter of fruit can be changed. When we did not fertilize the soil, the usage of NEVIROL decreased the fruit mass (it could reach 99.4 %) and the fruit diameter (on 98.3%). In the case of fertilization, the NEVIROL usage improved the fruit mass and diameter, but it was not significant. However, the yield amount (in both kg/tree and kg/ha) has increased remarkably. Due to fertilization the yield amount increased by 21.5% at Idared. For the other variety (Jonathan) the rate of yield increase was only 15.6%.

Table 4. Effects of NEVIROL 60 WP application on the pollination and fruit quality at two apple varieties, Idared and Jonathan

Experiment	Tretaments	Varieties	Pollination (%)	Fruit mass (g)	Fruit diameter (mm)	Yield amount (kg/tree)	Yield amount (t/ha)
Controll	Without	Idared	11.8	176	76.1	28.5	94.9
		Jonathán	10.2	140	70.5	25.2	84.2
	Fertilized	Idared	12.1	179	76.9	31.2	103.9
		Jonathán	10.9	143	71.3	26.8	89.3
Nevirol 60 WP	Without	Idared	14.5	175	74.8	30.9	103.0
		Jonathán	11.5	138	68.4	26.8	89.3
	Fertilized	Idared	15.2	179	77.2	34.6	115.3
		Jonathán	13.4	144	71.5	29.2	97.3

Grape

The other examined fruit species was the grape. We have chosen two traditional varieties for the NEVIROL usage. One was Muscat Ottonel and the other was Olasz rizling. The results can be seen in Table 5.

Table 5. Effects of NEVIROL 60 WP application on the pollination of two grape varieties, Muscat Ottone and Olaszrizling

Experiment	Treatments	Varieties	Number of flowers (number/ bunch)	Number of set berries (number/bunch)	Average set fruit (%)	Yield amount (t/ha)
Control	Without	Muscat Ottonel	298.5	118.6	39.7	12.9
		Olasz rizling	216.7	96.7	44.6	9.43
	Fertilized	Muscat Ottonel	299.1	128.4	42.9	13.1
		Olasz rizling	212.6	97.4	45.8	9.65
Nevirol 60 WP	Without	Muscat Ottonel	305.4	176.8	57.9	13.9
		Olasz rizling	214.3	112.3	52.4	10.3
	Fertilized	Muscat Ottonel	307.6	191.2	62.2	15.7
		Olasz rizling	211.5	118.6	56.1	11.9

In Table 5 it is demonstrated that there is not a big difference between the two grape varieties under normal circumstances, i.e. if we do not use NEVIROL. As a result of NEVIROL usage – without any fertilization – the number of set berry (average fruit set) has changed significantly. The rate of increase for variety Muscat Ottonel reached 29.2% (18.2%), while for Olasz rizling it was 16.1% (7.5%). When further fertilization was applied, the NEVIROL 60 WP had a great effect on yield amount and the number of set fruit. In the case of Muscat Ottonel the increase in the number of set fruits reached 49.3% and the rate of increase for yield amount was 19.8%. The reason of the differences is that many set berries do not develop into a matured berry till the harvesting date. We can get a similar result for Olasz rizling.

Cucumber

The third studied plant was cucumber. The NEVIROL usage results are shown in Table 6. The N-phenyl-phthalamic acid has the greatest influence on the size of the cucumber. The cucumber's price mainly depends on its size. There is an inverse relationship between prize and size. The NEVIROL can help to reach small-sized cucumbers. In the case of variety Barbara F1, it increased the rate of 3-6 cm sized cucumbers by 11% and the rate of 6-9 cm sized cucumbers reached 13.1%, while the lower priced, big-sized (9-12 cm) cucumbers' rate decreased by 20.1%. The reason for the phenomena is the increased number of cucumbers. Not only the small-sized cucumbers' number was higher in the fertilized treatment, but also that of the normal-sized ones. For variety Profi F1, we have to take into consideration the intensive growing rate of yield amount, which could reach 26.8% under fertilized circumstances.

Table 6. Effects of NEVIROL 60 WP application on yield quality and pollination at two cucumber varieties, Barbara F1 and Profi F1.

Experiment	Treatment	Varieties	Quality distribution in the percent of mass production			Percent of standard yield (%)	Yield amount (t/ha)
			3-6 cm	6-9 cm	9-12 cm		
Control	Without	Barbara F1	7.4	42.7	45.8	95.9	12.6
		Profi F1	8	41.6	48.1	97.7	13.3
	Fertilized	Barbara F1	7.3	42.7	46.4	96.4	13.8
		Profi F1	8	43.5	46.3	97.8	14.9
Nevirol 60 WP	Without	Barbara F1	7.6	44.6	43.5	95.7	13.9
		Profi F1	8.1	45.3	44.4	97.8	14.2
	Fertilized	Barbara F1	8.1	48.3	41.4	97.8	15.6
		Profi F1	8.4	46.6	43.1	98.1	18.9

Tomato

The research results in tomato by using NEVIROL can be seen in Table 7. For both varieties, the rate of set fruit and standard-sized yield have increased. We found a decreasing tendency in average fruit mass when we did not fertilize, although the yield amount slightly increased in this case. The fruit remained small-sized, the rate of saleable fruits decreased. In the fertilized case, when the standard yield amount was high, the yield amount increased in both cases. The rate of increase by NEVIROL usage could reach 7.6% for Delta F1 and 12.2% for Robot.

Table 7. Effects of NEVIROL 60 WP application on yield quality and pollination at the two tomato varieties, Delta F1 and Robot

Experiment	Treatments	Varieties	Average set fruit (db/bunch)	Standard yield (%)	Average mass of fruit (g)	Yield amount (t/ha)
Controll	Without	Delta F1	5.2	98.8	80	22.9
		Robot	7.8	98.2	64	50.1
	Fertilized	Delta F1	5.2	99.1	83	25.1
		Robot	7.9	99.1	68	53.4
Nevirol 60 WP	Without	Delta F1	5.4	97.3	78	25.2
		Robot	8.5	98.1	62	55.2
	Fertilized	Delta F1	6.5	99.1	84	27.0
		Robot	9.1	99.3	68	59.9

It can be proved that with the N-phenyl-phthalanic acid, the agent of NEVIROL 60 WP, we can improve the number of pollination and yield amount. The different species and varieties have different reactions to the usage of NEVIROL. After the usage, the level of fertilization has to be taken into consideration. Under low nutrient supply, the mass or diameter of the fruit will decrease, the yield will be broken up into little bits.

The N-phenyl-phthalanic acid as a regulator is not a substitute for the main elements of fruit and vegetable production and plant protection, if these are available for the production the favourable effect of the usage can be experienced.

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Summary

EFFECT OF N-PHENIL-PHTALANIC ACID (NEVIROL 60 WP) ON QUANTITATIVE AND QUALITATIVE PARAMETERS OF SOME HORTICULTURAL PLANTS

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At four plant species (apple, grape, cucumber, tomato), the influence of N-phenyl-phthalanic acid (NEVIROL 60 WP) has been studied on yield formation and yield quality by the authors.

The research results show that by using NEVIROL 60 WP we can improve the number of fruits, and the number of berries in a bunch. As a result of this process, the yield amount will increase.

NEVIROL has a great effect on the improvement of yield amount, especially under good nutrient supply. There is quite a big difference between the reactions of different species (for example, at those grape varieties, which have a loose bunch structure, the effect of NEVIROL is more favourable than at others). Before using NEVIROL, we have to take into consideration that the increased yield needs higher nutrient supply, otherwise, fruit mass and diameter will decrease, the yield will be broken up into little bits. In the study, the most favourable effect of NEVIROL 60 WP was detected under additionally fertilized conditions by the authors.

POSTER SESSION ENTOMOLOGICAL & ECOLOGICAL GROUP

NEW SPIDER MITE PEST IN THE VINEYARDS OF NORTH HUNGARY: GARDEN SPIDER MITE [*EOTETRANYCHUS PRUNI* (OUDEMANS, 1931)]

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The common pests among the mites of the vineyards in BAZ county are the grape erineum mite [*Colomerus vitis* (Pagenstecher)] and the grape rust mite [*Calepitrimerus vitis* (Nalepa)] from the *Eriophyidae* family; the two-spotted spider mite (*Tetranychus urticae* Koch) and the European red mite [*Panonychus ulmi* (Koch)] from the *Tetranychidae* family. Their damage can be detected generally every year, but its measure is different, up to the species and locality of the vineyard.
The garden spider mite (*Eotetranychus pruni*) was found in Hungary for the first time in the vineyards of BAZ county in 2000. Recently, the frequency of symptoms and the measure of infestation are more and more characteristics to the vineyards here. This spider mite species occurs almost on all of the cultivated vine varieties in the entire historical grape growing regions of BAZ county, especially in Hegyalja and Bükkalja.
Distribution and host-plants of the garden spider mite in the world:
Hungary – It is known from plum and apple trees of cultivated and abandoned fruit gardens as well (Bozai, 1971) and from purple crab and sloe (Ripka, 1998).
Bulgaria – in several regions, on vine (Balevski, 1980 cit. Schruft, 1985);
Greece – on apple, cherry, plum and peach (Papaioannou-Souliotis *et al.,* 1994);
Georgia – on ash-tree (Zajceva *et al.,* 1983);
The Unites States, England, Germany **–** on maple, horse chestnut, plum and grape (Jeppson *et al*. 1975, Baker & Tuttle, 1994);
The former Soviet Union – on plum, vine, apple, hazel and horse chestnut (Mitrofanov et al., 1987).

Description of the developmental stages:

Egg: Nearly spherical, width 0.01 mm diameter, surface plain, colouration yellowish-green
Female: Body oval, colouration yellowish-green, length of the body 0.32 mm, width of it 0.15 mm. Female has two lance-shaped eyes. Dorsal bristles with fine setae; four palpal segments
Male: Body oval, elongated, the same colouration as the female. Length 0.22 -, width 0.12 mm
Life cycle: The fertile female overwinters in the split of the vine stock or under the bark.
The egg laying begins after 3-4 days long matural feeding in spring, to the underneath of the lower leaves, close to the midrib or to the veins. The eggs are covered with fine strands of webbing.
Embryonic period is 15.4 days on 23 °C (Sepasgosarian, 1956).
The female lives from 38 to 53 days and average egg production is 37 (Sepasgosarian, 1956).
Depending on the ecological factors, 4-5 generations develop until the end of the vegetation period.
Hibernation starts at the end of August or the beginning of September.
Noticeable mortality can be observed only below minus 16.8°C (Sepasgosarian, 1956)

Observations

Detection of garden spider mite is possible only with a microscope, at least with 25x magnification, respecting the small size of this species. Developmental stages can be observed on the underneath of the leaves, closely to the midrib and veins.

Symptoms

The first symptoms appear on the leaves of the lower leaf floor, close to the ground and to the vine-stock, with a pale yellowish spot near to the midrib or to the veins, on the upper surface too. Feeding of the mites results reddish spots on red wine grape varieties. After this initial symptoms, the extension of these patches increase. Frequently, necrotic spots as a result of sun-ray are produced. Finally, the whole surface of the lower leaves wither, turn to yellowish-brown and the damage spread to higher floors of the vine-stock. If the population density is high, the deformation of the leaves can be observed. Complete defoliation of the lower leaf floor may follow when the mites are absolutely not, or not well controlled.

Economic importance

Garden spider mite is considered to be one of the most economically important spider mite in vineyards of BAZ county.
Due to the warming up of climate, shortcoming of the applied pest management and to the false determination of symptoms (are known as beginning of some fungal infection or lack of nutritional elements (e.g. magnesia) by the most of the vineyard owners), the intensity of the infestation and the extension of the infested area is increasing more and more.

Control

Out of the inspected acaricides, good result was given by pyridaben and amitraz active ingredients, especially when these were used at the beginning of the infestation.
It is necessary to start the control of this species in early season, at the appearing of the first symptoms (at the middle or end of May).When higher established population was observed (second part of June), only a very slight biological effectivity was provided by all of the inspected acaricides.
The spray should cover the underneath of the lower leaf floor and rather small spray drops should use in order to respect the size of the mite.

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ORNAMENTAL WATER-PLANTS AND THEIR PESTS IN HUNGARY

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The ornamental water plants are worldwide going to be more and more popular. This statement can be verified also by a plenty of special works and international expositions.
On the hilly countries of Hungary, the settlements developed a lot of dams across the valleys and made use of some abandoned mine outflows. Local leisure centres, fish ponds and recreational areas came into existence by the co-operation of inhabitants.
After changing regime, the social polarization resulted in prosperity of new type as the garden ponds became symbols of the upstarting. On a lower level, the aquaria appeared as flat ornaments. The equipments of these "mini-biotopes" are the ornamental plants and animals (fishes and turtles).
On the new biotopes, some new elements of flora and fauna appeared. These water-ecosystems must not be left alone. By means of implanting some attractive wild, semi-wild or cultivated water-plants, the new plant community could be regulated by the man.

Literature

Some species of the reed, sedge, rush and bulrush are gradually but independently appearing in the cold-watered lakes, creeks and backwaters (Fischl et al., 1998). The plant community can be accelerated and coloured by the human interventions. The *Lythrum* spp., *Potamogetan* spp., *Trapa natans* etc. are semi-wild water plants attractive and wantless which would settle down and multiply independently sooner or later (Botta, 1987; Krizsán, 1997). Some species of them will be presented briefly as follow.
The water marrow (*Nuphar lutea*) is denominated of its yellow flowers and characteristic fruits. It can be found in the standing waters and slow canals everywhere in the country. An ornamental water plant of the Kis-Balaton and the backwaters of Danube and Tisza.
Among the Lotus species, the perennial *Nymphaea alba* can also winter in the deep or shallow marshes of the flat areas of Hungary. It blooms from June to September continually. The petals of the opened flowers (10-15 cm in diameter) are white as snow, genital leaves yellowish. It can easily be

settled on sunny parts of some garden ponds. It grows rapidly and blooms abundantly when supplied with peat nutrition (Tuba – Bíró, 1987, Simon, 1992).
The ornamental water-plants which are Mediterranean by origin require a level of warmth above 20°C. The beautiful queens of the warm biotopes are the big-flowered *Nymphaea* species. These plants are the main ornaments of our warm ponds or pools because of their big round leaves, pretty flowers and perennial manner of life. The red water-lily (*N. rubra*) has from Félixfürdõ been settled into the lake of Hévíz by Lovassy Sándor in 1898. Since that time, a plenty of species and variants of several colours (yellow, blue, pink) are there in the lake and outflows.
The popularity of the water-lily in Hévíz is shown above all by the fact that this ornamental water-plant has an excellent place on the town-shield. Within cultural programmes, water-lily festivals have also been organized in Hévíz recently every year.
The Indian lotus (*Nelumb nucifera*) is a holy plant of India. It was one of the main adornments of the ponds of Pharaohs in the ancient Egypt. It is a horticultural secrecy till now, how the individual plants of diverse colours and appearances to be selected (Botta, 1987). Some exotic specimens of these plants are to be seen in the botanic garden of Szeged. Their leaves and flowers as well as fruits are adornments of the gardens.
Some later, after having the ornamental plants been settled, the pest also appear. As Linné's eternal truth says: "Every being has its own enemy hunting it forever".
The damaging pests of water-plants have by the researchers been neglected up today. There are only few exceptions: the damages on the reed were investigated by Vásárhelyi (1995), Fischl et al., (1998) and Bürgés et al., (1998) minutely. The damaging pests of water-lilies were noticed abroad by Sorauer (1954), Balachowsky (1963), Pape and Hemer (1964). The manner of life of the pests of the warm and cold watered biotopes in Hungary as well as the control possibilities are mentioned in the works of Bürgés – Horváth (1998a,b), Bürgés et al., (2000).

Materials and Methods

The investigations began 8 years ago. Our surveying have continually been done in warm ponds and outflows (Hévíz, Zalakaros, Kehida, Kincsesbánya, Félixfürdõ, Püspökfürdõ) as well as in cold-watered creeks, rivers and backflows (Mártély), mine inflows (Pötréte) and the Kis-Balaton.
Our main method is, by diagnosing the damages and diseases, the "plant individual survey". The grass net is an useful instrument for collecting. The less frequent pests will be brought up in hygrostats under laboratorial and free-field conditions. The observations on manner of life are done by tent

isolators and running instrument.

Results

By our investigations, 12 damaging pests of the ornamental water plants mentioned above were observed. From among the seven herbivore pests of the water-lilies, the *Galerucella nymphaea* and the *Rhopalosiphon nymphaeae* are the most important ones. The individual density and the number of generations of these species in the warm-watered spa make the regular control necessary. For this reason, the species mentioned above have to be presented more minutely.

Galerucella nymphaea L.

Description:
Imago: 6-8 mm, elongated oval yellow-brown sleek-haired body, flat wingtops. Mottles on wingtops. Feet and feelers darker.
Eggs: Spherical, opal-white, surface reticulated. Bunches of 10-15 pieces on the plant part emerging out of water surface.
Larvae: Young larvae black-green, after sloughing light brown. Larva after developing 8-10 mm, 3 foot-pairs, head capsule chestnut-brown, body elongated.
Pupa: Orange-yellow free pupa adhering to the surface of the leaf.
Spreading:
Spread but one of the less frequent species in Hungary. To be found on lake shores, marshes, rivers, canals or on water plants. Surveyed by us in warm-watered lakes and outflows as well as in the peat-mine waters of Pötréte, the river Marcal, the backflows of Tisza (Gyirmót, Tiszaug, Csépa) or on the water-plants of the Kis-Balaton. The species is also well-known by the gardeners caring for water-plants (Kincsesbánya, Tata, Háromfa, Balatonfüred) because of its damages.
The species has numerous host plants. It occurs primarily on water-plants which are floating on water surface, on (secondarily) on the wet land-plants. It shows a preference for the water-lily and the water-marrow, but also damages the *Potomogeton natus*, *Sagittoria sagittifolia*, *Rumex hydrolahum*, *Polygonum amphibium*, *Comarum palvistre*. Our surveys show that it damages also *Nymphaea rubra*, *N. marliaceae* var. *chromatella* and *N. coerulea*. The last species is the least infected one within the stock of Hévíz and this fact could be considered as a phenomenon of somewhat resistency.
Damage: The adults and the larvae are chewing at the surface of leaves.

The adults mostly pell circular polyeder surfaces on the leaves floating upon the water while the larvae gnaw shorter or longer canals on them. Having the epidermis got damaged, the leaves begin to become rotten and brown after a couple of days, losing decorative value. Some new leaves grow on the plants by intensive sprouting but these will be smaller and less attractive.
Developing cycle: In a cold-watered biotope the species has two generations. The adults winter on the shores among the fallen leaves. Our surveys show that the species has 3-4 generations in the warm lake of Hévíz every year. Elsewhere no investigations were done. The adults settle from the fallen leaves near to the water in early April. At first, they are only on the shores to be found. Later, as the air gets warmer, they mostly fly to the plants growing in the lake. After a couple days' feeding the adults copulate, then the females lay the groups of eggs on the surface of leaves. Our surveys by isolators show that a single female lays 80-120 eggs for a lifetime. She does it not at once but putting in some days' pauses.
The embryonic developing lasts 5-10 days. The larvae hatched out of the same group begin to feed scattering on the surface of leaves. During their development period four scales were observed thus they slough three times. The ripen larva, when finished feeding, becomes a pupa on the leaf. After a week, the new adults appear and soon begin to feed. The damages caused by them are by far not so serious than those of the larvae. The adults live long thus the generations fuse together.
Control: From among the preparations, *Bacillus thuringiensis* var. *tenebrionis* (Novodor FC) is allowed.

Rhopalosiphon nymphaeae L.

Its form, colour and size are mostly similar to those of *Aphis pisi*.
Host-plant circle is still unknown.
Damage: When the colony is populous, the leaves wind themselves up emerging out of the water, become withered and faded losing their ornament.
Developing cycle: It has 6-8 generations, the eggs winter on shrubby plants. The manner of life of the species is to be continued.
Control: The Vektafid-A containing paraffine oil is separately allowed. The spa of Hévíz and the natural water surfaces are protected to a greater extent, thus the control measures mostly encounter difficulties. The plant parts over the water surface were cut down when infected strongly. After placed on the shore, the biomass infected was covered with black plastics. The pests were killed by the heat becoming close under the plastics.
It is to be noted that the water-lily species have an excellent capacity for sprouding. They produce natural leaf-changes 4-5 times a year.

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Summary

ORNAMENTAL WATER-PLANTS AND THEIR PESTS IN HUNGARY

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The ornamental water-plants are worldwide as well as everywhere in the country going to spread. There are several pests and enemies of these plants which are practically unknown. Some of these are dealt with in this study.

Two important leaf-damaging pest species of the water-lilies (*Nymphaeae* spp.) and the water-marrow (*Nuphar lutea*) will be treated of as follows namely (*Galerucella nymphaea* L.) and (*Rhopalosiphon nymphaeae* L.). Because of high individual density and number of their generations in the warm-watered spa of Hévíz and its outfalls, a regular control is necessary.

LIGHT-TRAP EFFECTIVENESS DEPENDING ON THE PÉCZELY'S AND HESS-BREZOWSKY'S MACROSYNOPTIC WEATHER SITUATIONS

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In special literature light-trap effectiveness is interpreted differently by a number of authors.

The number of individual insects collected

In this sense effectiveness can be modified by the following factors:

- The physiological states age and sex of insects.
- The construction of trap, the wavelength and intensity of light applied placement of trap in a given environment (composition of flora in the surroundings, its phenological state, disturbing lights, altitude over sear level, etc.).
- Meteorological and cosmic factors (weather elements, macrosynoptic weather situations, atmospheric electricity, solar activity, ionospheric disturbances, environmental illumination, interplanetary magnetic field, moonlight as well as in close connection with it distance of insect attraction, and gravitational potential caused by celestial bodies, etc.).

Several authors have dealt with the above issues. As it is now not our duty to survey literature in more depth, we only refer to our monograph (Nowinszky, [ed.], 1994) which deals with the modification effects of meteorological and cosmic factors in details.

Collection area of light-trap

It signifies that distance, from which the insect species with varying vagility fly to the light-trap. In this sense a number of researchers have approached the concept of effectiveness.

The ratio of individual insects caught from those attracted by the light-trap

Only a smaller quantity of insects getting into the vicinity of light-trap, are actually caught by the trap, about 20 % (McGeachie, 1988). With this kind of interpretation of effectiveness only few researchers have dealt with, although the thorough examination of the topic would be extremely important for plant protection forecasting.

The ratio of individuals caught from the species present in the vicinity of light-trap

In literature we have not found the interpretation of light-trap effectiveness that we are going to present in the followings, neither in Hungarian nor in international literature. As a mater of fact it is not easy to determine this ratio, as one has first define what is understood under species "present". It is well known that mobile and vagile species can fly into the light-trap that do not live in the given area, but are coming from a longer distance. Consequently the individual insects caught are made up from the individuals that have developed on plants in the vicinity of the trap and individuals arriving from a longer distance, e.g. migrants. The local insect population can anytime added by immigrant individuals, which cannot be unambiguously separated from the locally developed ones. (Mészáros, 1987-88). The vigility of the various species is different thus they can get into the vicinity of the light-trap from various distances, even in case of identical environment illumination. In the daily entrapped material there appear individuals of such species on one hand that do not belong to the population living in the direct surrounding, but arrive from longer distances. On the other hand, similarly to the trapping procedure based on other luring methods. Those individuals of local insect populations do not appear in them that do not react to the trap stimulus for whatever reason. Consequently we understand under species "present" those individuals which stay at a given time at such a distance from the light-trap so that they can react to trap stimulus, irrespective of whether the do it actually or do not effected by any inhibitions. Thus the term "present" means in our opinion an actual distance that is differing species by species.

We have set the following objectives for our study:

- Elaboration of a method based on light trap collection data for the calculation of number of species present in the vicinity for any day of the year, and based on this determination of number of aspects, their appearance in time and the time duration of their existence, as well as the effectiveness of the trap,
- Elaboration of a method to understand the relationship between trap effectiveness and weather situations,
- Comparison of Péczely's and Hess-Brezowsky's macrosynoptic typologies for determining which one of them is more reasonable to be used in case of agrometeorological processing of light-trap data.

Materials and Methods

Data were collected and obtained from the material of forestry light-trap in Szombathely, one of the uniformly equipped national Jermy-type light-trap network. The Jermy-type light-trap is a modified version of the Minnesota-type from which the gatherer sheets are missing. Its light source is a normal bulb displayed at 2-m height, with colour temperature of 2900 ºK, the killing substance is chloroform. The traps are operated by the research institutes and plant protection sites from April 1st to October 31st, the forestry traps are operated the whole year round, irrespective of weather, sunset or sunrise conditions all day from 18 PM to 4 AM. The traps do not operate on days when temperature does not rise over 0 ºC, or if the area is covered with snow. Insects get the whole night into a single collecting glass. The results of collection per night mean one data.

The light-trap chosen for our investigation was operated in Szombathely between 1961-1970 within the premises of the Kámon Arboretum. The complete Macrolepidoptera material of this observation site was used to examine light-trap effectiveness. Collection period species collected and number of swarming are displayed in Table 1.

Table 1. Light-trap collection periods as well as the number of caught species and swarmings

Years	*Collection periods*	*Number of species*	*Number of swarmings*
1962	March 05 – November 21	343	435
1963	March 08 – December 03	349	472
1964	March 23 – December 19	354	463
1965	March 14 - December 21	205	242
1966	February 02 - December 02	153	191
1967	February 03 - November 19	261	312
1968	February 20 - November 26	296	418
1969	March 13 - November 27	316	427
1970	February 03 - November 30	323	437

Data related to macroscopic typology and the detailed description of the situations can be found in the papers of Károssy et al. (1994) and Nowinszky et al. (1994).

Trap effectiveness was calculated on every day of the 9-year period from the Macrolepidoptera material of the light-trap Szombathely. The numbers of individuals of the respective species were not considered on a daily basis, it was only examined whether certain species was present on a

particular day. Data on more-generation species were processed separately, according to generations. On the other hand if between the swarming time of two generation's vagile or migrating individuals between the swarming period of two generations could be easily observed, these were considered as independent generation. If the two generations were not to be separated unambiguously from each other, the procedure used with one-generation species was followed.

The trapping data of the first sample of a given generation is called appearance, and the day following trapping data of the last individual is called disappearance. The frequency of appearance and disappearance of all generations of species were summarised day by day, then it was cumulated and illustrated. The difference between the cumulated appearance and the disappearance was calculated. This way we obtained the number of species present in the surrounding of the trap as a function of time. The number of species trapped daily was determined from the light-trap record and displayed with the species present.

The individual species of course appear and disappear continuously, thus the aspects following each other cannot be sharply distinguished. We have determined the division lines of aspects through the following procedure: the total amount of appearing (A) and disappearing (D) species was calculated day by day and illustrated and the most periods of most dynamic changes identified. These were compared with the rising curves of (A) and (D) as well as with present (P) curves and the approximate time data of aspect changes could be read. Finally ratio of entrapped individuals compared with those present in the vicinity was calculated in percentages. This result is what we considered to be the effectiveness of the trap (E). In the followings the daily effectiveness data for the 9 years were jointly handled, but sorted according to aspects and correlated according to the Péczely's and Hess-Brezowsky's macrosynoptic type codes numbers for the days in question and averaged. Then we controlled the significant deviation of averages from the average value of the aspect.

Results

The effectiveness of the light-trap in Szombathely is correlated with the Péczely's and Hess-Brezowsky's macrosynoptic weather situations, according to the aspects and illustrated in Table 2 and Table 3.

Table 2 The effectiveness of the light-trap in connection with the Péczely's macrosynoptic weather situations

Péczely's situations		*Spring aspect*		*Early summer aspect*		*Late summer aspect*		*Autumn aspect*		*Winter aspect*	
		E %	*N*	**E %**	*N*	**E %**	*N*	**E %**	*N*	**E %**	*N*
mCc	(1)	**20.3**	53	**18.9**	56	**26.0**	38	37.5	6	0	4
AB	(2)	**12.7**	31	**21.5**	34	**26.7**	46	31.4	12	22.2	1
CMc	(3)	**17.4**	36	23.9	24	**21.6**	21	10.9	8	2.0	6
mCw	(4)	**33.7**	98	**28.0**	71	29.0	71	**33.1**	51	14.6	15
Ae	(5)	**34.2**	74	**30.3**	34	**34.7**	100	31.0	88	3.7	8
CMw	(6)	**19.2**	60	**15.7**	33	**23.9**	38	**22.3**	46	0	6
zC	(7)	**35.5**	52	25.4	18	26.4	31	**46.3**	22	19.9	7
Aw	(8)	**21.3**	107	27.6	131	**26.0**	143	**23.8**	71	7.4	13
As	(9)	**35.9**	27	20.4	20	29.0	21	**36.8**	31	25.0	4
An	(10)	**19.1**	69	26.0	74	**32.5**	103	**35.0**	41	0	1
AF	(11)	16.8	15	**30.1**	24	**32.8**	35	16.2	6	0	2
A	(12)	**38.3**	27	**34.9**	47	**32.9**	85	26.6	72	1.4	10
C	(13)	23.3	13	26.5	7	25.9	11	9.5	5	33.3	3
Averages:		*25.8*		*25.9*		*29.4*		*29.4*		*9.4*	

Notes: E = efficiency of light-trap (%). Bold numbers sign if the significance levels of average effectiveness values are higher than 95 % as well as the deviation characteristic for a given aspect.

Discussion

The almost up-to-date Péczely typology and the regular publishing of codes allow both the continuity of the related entomological research and the processing of the latest observation data. Based on the former examinations exploring the effect of weather factors as well as trapping results correlated with the individual Péczely-type situations one can determine those weather situations that are favourable or unfavourable from light trapping of insects.

If we compare the typology considering the surface baric field as well as the Hess-Brezowsky's typology based on 500 bar atmospheric levels, it seems proved that light trapping effectiveness in Hungary can be more closely related to Péczely's typology relating to the lower levels of planetary border stratum.

Péczely's macrosynoptic weather situations are not only valid from the point of view of climatologic typology but they are also apt for agrometeorologic research purposes as well. Elaboration of similar typologies for other geographic regions seems also reasonable.

Table 3. The effectiveness of the light-trap in connection with the Hess-Brezowsky's macrosynoptic weather situations

Hess-Brezowsky's situations		*Spring*		*Early summer*		*Late summer*		*Autumn*		*Winter*	
		aspects									
		E %	*N*	*E %*	*N*	*E %*	*N*	*E %*	*N*	*E %*	*N*
Na	(11)	17.1	10	31.4	4	29.3	4	-	-	-	-
Nz	(12)	13.2	20	33.9	15	29.2	11	32.2	6	0	1
HNa	(19)	18.3	15	31.9	21	31.3	9	58.7	9	-	-
HNz	(20)	26.2	10	24.5	14	31.9	12	16.4	4	-	-
HB	(18)	8.3	13	15.0	12	16.9	16	36.2	8	11.1	2
NWa	(13)	26.6	7	36.8	11	17.0	12	-	-	-	-
NWz	(14)	**19.2**	56	32.8	13	25.7	40	26.2	9	11.4	3
TRM	(27)	**9.1**	28	**19.1**	26	24.0	24	23.4	18	3.2	9
Wa	(1)	37.7	13	**29.1**	42	30.0	36	**21.5**	39	-	-
Wz	(2)	**35.5**	119	24.8	66	29.3	109	28.2	87	9.6	23
Ws	(3)	**43.1**	22	22.8	9	30.1	27	58.3	12	13.3	15
Swa	(7)	**36.7**	20	36.0	15	32.5	27	24.6	24	-	-
SWz	(8)	**35.4**	43	32.8	13	31.5	30	25.7	29	0	6
TRW	(28)	**31.0**	31	22.5	28	30.7	47	34.6	23	-	-
Ww	(4)	15.1	28	26.0	24	29.6	15	50.0	2	0	3
Sa	(5)	38.6	13	16.7	1	31.8	10	33.2	24	-	-
Sz	(6)	35.7	8	-	-	40.8	4	36.7	13	-	-
TB	(26)	26.8	11	21.1	17	**38.5**	27	23.4	18	-	-
SEa	(9)	14.6	12	32.2	6	17.2	7	12.5	8	-	-
SEz	(10)	40.0	2	32.8	3	25.0	3	-	-	-	-
HFz	(22)	27.8	19	25.1	23	28.7	26	40.1	16	-	-
HNFa	(23)	29.9	35	21.1	11	26.5	8	-	-	0	3
HNFz	(24)	-	-	26.5	14	31.1	17	59.0	5	28.6	7
Nea	(15)	16.3	9	21.3	22	18.9	7	0	2	0	3
Nez	(16)	22.6	7	22.5	21	25.3	18	0	2	-	-
HM	(17)	**19.7**	36	**31.5**	31	30.4	70	**24.8**	67	-	-
BM	(30)	30.2	30	27.8	19	31.9	69	**21.4**	38	-	-
TM	(25)	**8.3**	33	21.1	17	24.8	12	37.8	9	-	-
Averages		*25.8*		*25.9*		*29.4*		*29.4*		*9.4*	

Notes: E = efficiency of light-trap (%). Bold numbers sign if the significance levels of average effectiveness values are higher than 95 % as well as the deviation characteristic for a given aspect.

There are more examinations required to explore the effect of the Hess-Brezowsky's macrosynoptic weather situations (valid for whole Europe) in Hungary as well. Irrespective of this their utilisation seems to be promising

for forecasting in plant protection. The Péczely-type macrosynoptic weather situations are probably easier to interpret and give a less ambiguous picture of weather conditions. Thus we recommend their application in researches that focus on insect ecology, serving the purposes of forecasting in plant protection in Hungary. Hess-Brezowsky's macrosynoptic types can in our opinion be utilised first of all in international research efforts, for example in controlling migrating insect populations.

The better understanding of the role of whether allows making better forecasts for plant protection. Thus the results of our work can contribute to the application of environment-friendly, effective and at the same efficient plant protection procedures. The application of our method allows to research the relation of life phenomena of insects with weather even in those cases, when measuring the individuals factors hits against difficulties. The collection data of the national light trap network are of invaluable scientific importance and can also be utilised in research of insect ecology and etology. The results of our method to determine effectiveness of light trap can be utilised in ecology, cenology and faunistic research.

Even from the data continually collected at an observation site over a period of year can serve for drawing conclusions as related to various taxons, supposing naturally that the research is not restricted to reporting the collection data of only a few insect species:

- The number of species present in the surroundings can be determined for any day of the year
- The number of aspects, their appearance in time and the period of their existence can be determined,
- The periods of rapid changes and relatively steady periods are recognisable,
- Trap effectiveness can be calculated indicating the percentage of species entrapped by the trap in a given period,
- The alteration of effectiveness is comparable by aspects, by taxons and by sexes can indicate their different demands against the abiotic factors.

The data obtained at the same observation site in different years, as well as the data gained at various observation sites for the same years can also be evaluated on the basis of the categories listed above. The differences manifested in the number of aspects, the time of their appearance and disappearance, the period of their existence, species diversity as well in trap effectiveness can be compared with data on climatic and weather conditions. Observation covering a number of years can indicated unfavourable changes occurring in the environment, for example the continuous decrease of species as a consequence of environment pollution. Thus the method

presented here can play a major part in research related to environment protection.

As a matter of fact, the method presented has the shortcomings that are connected with the peculiar features of light trapping. In addition one has to consider that the involvement of those species that are only detected for one or two days in trap material can lead to overestimation of trap effectiveness. Their swarming lasts obviously for longer time, but the method is not able to show the decrease of effectiveness on other days. On the other hand effectiveness is under-estimated if the swarming time of two subsequent generations is inaccurately divided from each other or if we consider the whole period of collecting a given species as one swarming because of the individual arriving from longer distances. Most of these potential failures can be avoided if one has been adequately prepared professionally, if phenology of the various species at the given observation site is studied in a thorough way, and if in case of processing fresh insect material, migrating individuals are singled out.

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Summary

LIGHT-TRAP EFFECTIVENESS DEPENDING ON THE PÉCZELY'S AND HESS-BREZOWSKY'S MACROSYNOPTIC WEATHER SITUATIONS

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The authors developed a method for calculating the number of species present in the vicinity of the trap for any given day of the year. Using this, we determined the number of aspects, the time and duration of their appearance, as well as the effectiveness of the trap. We studied the daily effectiveness with regard to the Hess-Brezowsky's and Péczely's macrosynoptic weather situations use up the data of Jermy-type light-trap operated at Arboretum in Szombathely. We took the code numbers from the Péczely's (1983) and Hess-Brezowsky's (1977) catalogues. According to the days of the calendar we summed up and then cumulated the frequency with which each generation of each species appeared and disappeared. The difference between the cumulated appearance and disappearance gives the number of species present in the vicinity of the trap. We plotted this, then determined when the changes of aspects occurred. Next we determined the number of species trapped daily. Finally we calculated the percentage of insects trapped in comparison to those present. This result is what we considered to be the effectiveness of the trap. The daily effectiveness data for the nine years sorted according to aspects and correlated according to the Péczely's and Hess-Brezowsky's macrosynoptic type code numbers for the days in question was averaged. We were able to determine the macrosynoptic situations favourable and unfavourable for collecting.

QUANTIFYING BIODIVERSITY IN ECOSYSTEMS BY GREEN LACEWING ASSEMBLAGES I. A VALUABLE METHOD TO DO (INSECTA: NEUROPTERA: CHRYSOPIDAE)

(Summary)

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Green lacewing samplings coming from eleven various significant biotopes were chosen to characterize the biodiversity they display. They point out different types of landscapes: Mediterranean and montane forest biotopes, two wet meadow-lands and a vegetable field in plain, one subspontaneous and three managed Mediterranean olive groves, a submediterranean calcareous slopes and an oro-mediterranean moorland. Three parameters were used to quantify biodiversity. They betoken: (1) the rough faunistical richness (Margalef's index), (2) the diversity as the relative importance of each collected species and the ratio between the total numbers of species and individuals (Shannon's index), and (3) the equitability or relative heterogeneity featuring the distribution of the species specimens and giving an idea of their dominance (Hulbert's index). To ascertain the values recorded on single samplings, we operated by the method of Bootstrap. We calculated a confidence interval (Box Plot) coming from 10,000 virtual samplings, simulated by randomly picked up pullings within a infinite population showing the same distribution of species than the original assemblage. A classifying process (Cluster Scatterplot) was then established in order to appraise the proximity of the different habitats.

The Shannon and the Hulbert's indices bear out the high sensivity of these parameters to the structure of studied assemblages. They evidence a conspicuous difference of the montane forest biotope: the number of rare species was fairly high, seven species were abundant mainly *Cunctochrysa albolineata* (48.1 %) and *Chrysopa pallens* (26.9 %). Reversely, in the scanty agroecosystem and also at a lower level in the sites strongly alterated either by agricultural farming practices or by climatic cues (inundation), the faunistical richness and the diversity were low whilst a strong dominance of the common green lacewings appeared. Mediterranean biotopes together with the wet meadow are characterized by an equitability always higher than

0.48, unless the number of collected species. The wild Mediterranean biotopes constituted by the maquis moorland, the typical Aleppo-pine forest and the calcareous slopes look like both rich and well-balanced biotopes, however better than the two insecticide-free olive groves. The analysis results in a diagrammatic typological approach of the biotopes.

Establishing average diversity indices together with their confidence intervals allows unambiguous comparisons between various chrysopid assemblages. By the way, one may characterize their plight relative to the more or less abundant and diversified occurrence of these polyvalent predators which are a valuable signal of a good ecological functionning within ecosystems. In farming advising, such an approach of checking, forecast and management is promiseful for all responsibles and advisors of future agriculture which must become and remain sustainable and respectful of the planet resources.

QUANTIFYING BIODIVERSITY IN ECOSYSTEMS BY GREEN LACEWING ASSEMBLAGES. II. AN EXAMPLE FOR EVIDENCING CHRONOLOGICAL CHANGES IN AGROECOSYSTEMS (INSECTA: NEUROPTERA: CHRYSOPIDAE)

(Summary)

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The ability of an agroecosystem to enter sustainable development needs control and mainte-nance in time of its good health, failing which they fall into disrepair. Owing to the availability of quantifying accurately biodiversity indices, we demonstrate here by an example the possibility of detecting biodiversity chronological changes during a short period by means of green lacewing samplings. Light-trap data of green lacewings captured from 1985 to 1994 in the vicinity of Bucharest (Romania) suggests that there have been alterations in the species assemblages. The main occurring species were those constituting the common green lacewings, i. e. the *Chrysoperla carnea* complex, plus *Chrysopa formosa* Brauer, *Chrysopa pallens* (Rambur) and *Chrysopa perla* (Linnaeus). They are present every year whilst several other casual species did not. Among the latters, *Chrysopa abbreviata* Curtis, *Chrysopa nigricostata* Brauer and *Cunctochrysa albolineata* (Killington) disappeared along time contrarily to eurytopic and more generalist species. *Chrysoperla carnea* (Stephens) *sensu lato* was almost double in its relative frequency when measured during a similar (summer) period, increasing from 38 to 72 %.

Three parameters were used to quantify biodiversity. They betoken: (1) the rough faunal richness (Margalef's index), (2) the diversity as the relative importance of each collected species and the ratio between the total numbers of species and individuals (Shannon's index), and (3) the equitability or relative heterogeneity of populations, featuring the distribution of the species specimens occurring in an assemblage and giving an idea of the dominance of the more abundant species (Hurlbert's index). To ascertain the values recorded on a single sampling in each case, we operated by the method of Bootstrap. We calculated a confidence interval (Box Plot) coming from 10,000 virtual samplings, simulated by randomly picked up

pullings within an infinite population showing the same distribution of species than the original collection. A classifying process (Cluster scatterplot) was then established in order to appraise the proximity of annual values.
The present study did not show any significant decrease in the basic biomass of lacewings. The faunal global richness seemed not very perturbed, only lessening of about 9 % of its initial value. The diversity and equitability indices relative to each year manifested a rather high level of biodiversity for an agricultural environment. Nevertheless, they decreased significantly with time (parametric and non-parametric tests). They left about 45 %, meaning both together a loss in diversity. The progressive dominance of the ubiquist common green lacewings is attested by a logistic regression showing a good adequation. It constitutes a strong cue of alteration in the crop fields and a preliminary upsetting statement.

Key words: green lacewing, biodiversity, biodiversity indices, agroecosystem, light trapping, Romania.

DISCOVERY OF A LONG-RANGE CHEMICAL ATTRACTANT FOR LACEWINGS (*CHRYSOPERLA* SPP.)
(Summary)

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In the course of field trapping tests in Hungary originally aimed at capturing female noctuids, regular catches of green lacewings were observed in traps baited with phenylacetaldehyde, a well known attractant for several Lepidoptera. In subsequent trapping tests at several sites in Hungary and Italy, traps baited with one (lower dose) or three (higher dose) polyethylene bag bait dispensers loaded with this compound caught significantly higher numbers of green lacewings than unbaited traps, confirming beyond doubt the long-range attractivity of this compound. There was no significant difference between catches of traps with one or three dispensers. Both sticky delta and funnel traps baited with phenylacetaldehyde were capable of catching green lacewings and there was no difference between the performance of the two trap types. Funnel traps baited with three dispensers were used for monitoring the occurrence pattern of green lacewings throughout all the season with success. Captured specimens belonged mainly to the *C. carnea* species complex.

Until now, only a few substances were described as attractants for green lacewings. Some of these were tested by other groups to increase common green lacewings (*Chrysoperla carnea* s.l.) in arable crops with different results, which may in part be due to the difficulty to distinguish the effect of confounding factors (such as any competing odours from the crop, influence of naturally-occuring prey populations, weather conditions, and so on) under field conditions.

Chrysoperla carnea s.l. is a key-species in several crops for the control of a wide range of pest aphids, caterpillars, and other soft-bodied arthropods. The attractant discovered in this study showed approximately the same level of attraction towards female and male specimens of the common green lacewing whereas a wide range of plant volatiles tested by Dodds and McEwen (1998) showed a stronger intensity of attraction towards male specimens of *C. carnea*.

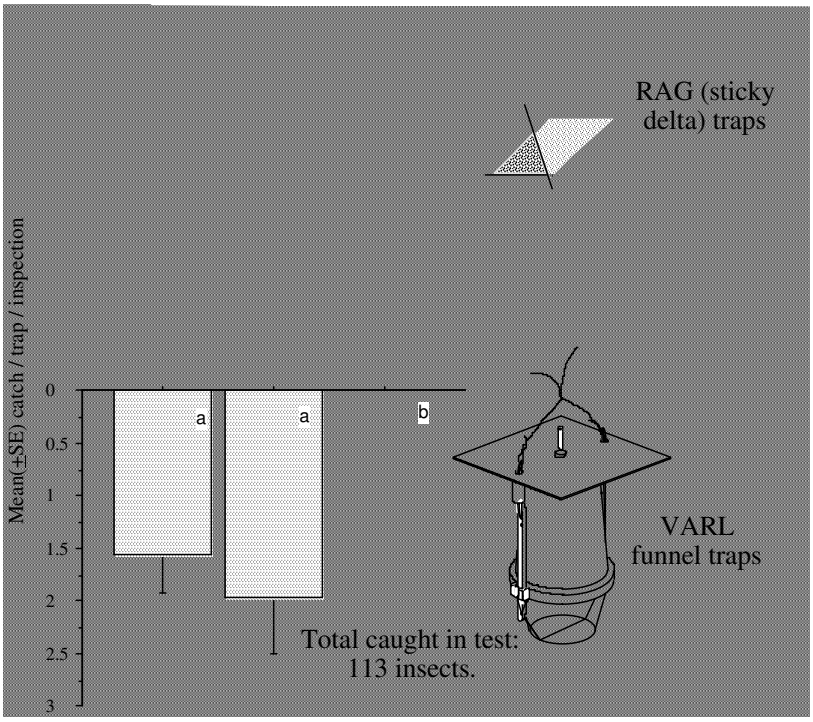

Fig. 1. Catches of green lacewings in sticky delta or funnel traps baited with phenylacetaldehyde vs. unbaited traps at Halásztelek, Hungary, May 9 - June 10, 2003.
All specimens captured belonged to the *C. carnea* species group (establishment of species identity of single specimens captured is underway). Significance: columns with same letter within one diagram not significantly different at P=5% by ANOVA, Games-Howell.

Further research would focus on the possibility of application of the new attractant on its own or in combination with previously known attractants for the study of lacewings in different crops.

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RESPONSE OF THE ANTIOXIDATIVE DEFENCE TO FLAVONOID QUERCETIN IN TWO POPULATIONS OF *LYMANTRIA DISPAR* L.

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Lymantria dispar L. a polyphagous herbivore is the most dangerous insect pest of forest and fruit trees. Its host range is estimated at more than 500 plant species from 73 families (Lance, 1983, Liebhold et al., 1995). The locust tree *Robinia pseudoacaccia* is a plant that the gypsy moth avoids as a food (Barbosa and Krischik, 1987). Locust tree leaves contain large quantities of alkaloids and flavonoids (Bagrbosa & Krischik 1987). Some of them may have toxic and prooxidant effects (Hodnick et al., 1986). Gypsy moth populations in locust tree forest are rare (Jankovic 1958). The ingestion of oxidizable flavonoids can exacerbate oxidative stress in herbivorous insects (Ahmad, 1992; Felton and Summers, 1995; Pardini, 1995). The flavonoid quercetin used in this experiment was chosen as test prooxidant plant allelochemical. Upon insect ingestion quercetin is metabollicaly activated by one-electron oxidation to a free radical (o-semiquinone) which in turn reacts with O_2 (oxygen) to generate $O_2^{\cdot -}$ (superoxide anion radical) and consequently H_2O_2 (hydrogen peroxide) and $^{\cdot}OH$ (hydroxyl radical) resulting in numerous destructive reactions in insect cell (Hodnick et al.,1986; Hodnick et al.,1989).

The cellular antioxidative defense of herbivorous insects includes the enzymes (superoxide dismutase-SOD, catalase-CAT, glutathione-S-transferase-GST, glutathione reductase GR, ascorbat peroxidase and dehydroascorbate reductase) and antioxidants (e.g. ascorbic acid, glutathione and α-tocopherol) that protect cells from oxidative stress (Ahmad 1992; Felton and Summers 1995; Pardini 1995). Considering that the gypsy moth population is present in the Bagremara (our experimental population) for more than 50 years (Sidor & Jodal, 1983), it is to a certain extent adapted to a locust-tree leaves diet (Peric-Mataruga et al., 1997; Lazarevic et al., 2002). Our previous results have shown that locust tree leaf diet lead to on increase in GST and SOD activities and GSH content as well as to a decrease in CAT activity in the midgut tissue. Fifty-year adaptation of the gypsy moth population to the unfavourable host plant in the locust tree forest have resulted in the changes of antioxidative defence (Peric-Mataruga et al., 1997). The adaptive changes of the constitutive expression

of the activities of antioxidative enzymes of the pest insects are very important component of their susceptibilty to insecticides (Gordon 1961).
The aim of this research was investigating the effects of artificial diet supplemented with the flavonoid quercetin (1.5%w/w) on the level of midgut tissue antioxidative defence: the activity of superoxide dismutase (SOD), catalase (CAT), glutathione-S-transferase (GST), glutathione reductase (GR) and glutathione content (GSH) in the 4th instar of the gypsy moth originating from oak and locust tree forest.

Materials and Methods

Egg masses of *Lymantria dispar L.* were collected from two localities (oak forest – "Bogovadja", locust-tree forest – "Bagremara"). During the winter the egg masses were kept at 4°C until May when they were transferred to a constant temperature of 23°C to hatch. After hatching the gypsy moth caterpillars were divided into the following four experimental groups:

- OC- caterpillars from the oak forest fed arteficial diet without quercetin
- OQ- caterpillars from the oak forest fed arteficial diet supplemented with quercetin (1.5% w/w).
- LC- caterpillars from the locust-tree forest fed arteficial diet without quercetin
- LQ- caterpillars from the locust-tree forest fed arteficial diet supplemented with quercetin (1.5% w/w).

The caterpillars (4th instar) were reared in plastic containers (2dl) at 23 °C and fed standard artificial diet for gypsy moth (O′ Dell et al., 1985) with or without quercetin (3,3',4',5,7-pentaxydroxyflavone, Sigma Chemicals Co., St Louis, Misouri). After the caterpillars were sacrificed the midguts were dissected on ice, washed several times with ice-cold physiological saline solution (0.9 % NaCl), midgut peritrophic membrane with content was removed and midgut were rinsed again with ice-cold physiological saline solution again. Midguts of 7-10 larvae were pooled by weight and homogenized in 0.25 M sucrose, 0.05 M Tris-HCl, 1mM EDTA pH=7.4 buffer (1:10 w/v) according to Rossi et al. (1983), and sonicated according to Takeda et al. (1982). For determination of the total amount of glutathione, part of the sonicated homogenate used to precipitate proteins with 5% sulpho-salicylic acid and the total amount of glutathione was measured after centrifugation at 5000 rpm for 10 min. The rest of the sonicated homogenate was centrifuged at 10500g for 90 min and the activities of SOD, CAT, GST and GR were determined in the supernatant.

SOD activity was determined according to Misra and Fridovich (1972). The method includes monitoring the degree of inhibition of adrenaline autooxidation in an alkaline medium in the presence of SOD. The enzyme unit was defined as the amount of enzyme inhibiting 50% of the control reaction and was expressed per mg protein.
CAT activity was determined by monitoring spectrophotometrically the degradation of a standard concentration of hydrogen-peroxide (Beutler, 1982) and was expressed as nmol H_2O_2 /min /mg protein.
Habig′s method (Habig et al., 1974) was used for determining GST activity. The unit is defined as nmol GSH /min/ mg protein.
GR activity was measured according to Glatzle et al., 1974, by monitoring spectrophotometrically changes of the amount of NADPH consumed for the reduction of a standard amount of oxidized glutathione (GSSG). The activity was expressed as nmol NADPH /min/ mg protein. All enzyme assays were performed at 30°C. The total amount of glutathione both oxidized and reduced was measured according to Griffith 1980 and was expressed per g wet midgut mass.
The statistical significance of the results was estimated by analysis of variance (Sokal and Rohlf, 1981).

Results

Activity of the superoxide dismutase in the midgut tissue of the larvae fed artificial diet supplemented with quercetin (OQ and LQ) was higher than in the control groups (OC and LC) (Table 1). Two way ANOVA confirmed significant effect of quercetin in a diet for the SOD activity (Table 2). This difference was more expressed in a oak population which were more sensitive to nutritional stress. SOD activity was higher in a control group from locust tree (LC) than in a control group from oak population (CO) (Table 1).
An artificial diet with quercetin was associated with a decrease of the gluathione-S-transferase activity regardless of the population origine (Table 1.). Two-way ANOVA revealed significant population and significant host-plant effects of the diet suplemented with quercetin on the GST activity in the midgut tissue of the gypsy moth caterpillars (Table 2.). The effect is more pronounced in oak adapted than in locust tree adapted population. Both populations showed the trend of elevated total amount of glutathione in the midgut tissue as a response to quercetin supplemented arteficial diet (but with no significance) (Table 1.).
Quercetin in the artificial diet did not change activity of the catalase and glutathione reductase in the midgut tissue of the larvae of bouth populations.

Table 1. Activity of antioxidative defence enzymes and amount of glutathione in the midgut tissue of 4th instar gypsy moth larvae originating from different populations and fed artificial diet supplemented with quercetin (1.5% w/w quercetin)

OC- caterpillars from the oak forest fed arteficial diet without quercetin
OQ- caterpillars from the oak forest fed arteficial diet supplemented with quercetin (1.5% w/w)
LC- caterpillars from the locust-tree forest fed arteficial diet without quercetin
LQ- caterpillars from the locust-tree forest fed arteficial diet supplemented with quercetin (1.5% w/w)

	OC	OQ	LC	LQ
SOD	10.77 ± 1.46	21.39 ± 1.78	16.01 ± 1.21	21.37 ± 1.49
CAT	88.9 ± 25.8	120.17 ± 20.0	99.91 ± 13.3	84.67 ± 1.65
GST	29.77 ± 2.17	17.62 ± 4.26	13.17 ± 3.65	10.46 ± 0.6
GR	1.35 ± 0.275	1.79 ± 0.29	1.73 ± 0.625	1.45 ± 0.425
GSH	0.83 ± 0.08	0.93 ± 0.12	0.73 ± 0.068	0.95 ± 0.065

Discussion

Due to its poliphagous nature, the gypsy moth is exposed to a variety of allelochemicals, some of which has prooxidant effect. The preference of gypsy moth caterpillars for host plants correlates negatively with the presence of flavonoids and alkaloids (Barbosa and Krischik 1987).

Upon insect ingestion quercetin can be metabolically activated by one-electron reduction to generate free radical species, which can further react with molecular oxygen to form the oxygen radical, superoxide (Hodnick et al., 1989).

Our results show that both oak and locust tree caterpillars fed on diet supplemented with quercetin have higher SOD activity than control groups (Table 1.). SOD is an antioxidative enzyme that catalyzes the dismutation of superoxide radical to hydrogen peroxide (Fridovich 1978). It is intersting that SOD activity in the midgut tissue was higher in a control group from locust tree population than in a control group from oak forest (Table 1). Superoxide dismutase is one of the most important components of the antioxidative defence against prooxidant effects of quercetin (Pritsos et al., 1988). This high constitutive SOD activity explains a potential of the gypsy

Table 2. The two-way analysis of variance (ANOVA) for the impact of population origin – P and types of diet (artificial diet and artificial diet supplemented with quercetin) – D on the levels of components of antioxidative defence in the midgut tissue of 4th instar gypsy moth

		P	D	PxH	Error
	df	1	1	1	17
SOD	MS	0.027	0.29	0.026	0.0093
	F	2.9	31.06 ***	2.84	
	df	1	1	1	17
CAT	MS	0.0017	0.0091	0.0855	0.043
	F	0.038	0.21	1.96	
	df	1	1	1	14
GST	MS	0.17	0.24	0.02	0.0111
	F	15.69***	21.37***	1.84	
	df	1	1	1	14
GR	MS	0.017	0.00027	0.0126	0.042
	F	0.401	0.0064	0.3	
	df	1	1	1	17
GSH	MS	0.0011	0.029	0.0083	0.0108
	F	0.105	2.712	0.767	

*P<0.05; **P<0.01; ***P<0.001;

moth population from the locust forest to survive at higher quercetin concentration in the artificial diet than oak population (Peric-Mataruga et al., 2001). As the gypsy moth had inhabited the locust tree forest for more than fifty years it is likely that a trophic process occured which is also manifested in the rise in SOD activity. The dismutation reaction catalysed by SOD results in the production of toxic H_2O_2 (Fridovich 1978). H_2O_2 is scavenged by another antioxidant enzyme catalase (Ahmad et al., 1987). Since there was no changes in CAT activity in the midgut of the treated groups of the gypsy moth, toxic effect of quercetine can be attributed to H_2O_2 mediated effect.

Our results showed that glutathion-S-transferase activity decrease if caterpillars from both populations fed diet supplemented with quercetin (Table 1). In insects GST is inportant in metabolic detoxification of

insecticides (Yu 1996), of allelochemicals from host plants (Yu 1993) in protect insects from the toxic effects of active oxygen species (Parkes et al., 1993, Zaman et al. 1994, Hodnick et al., 1996) and for the turning on the detoxifying enzymes enhancing the defense machinery, speeding the development of resistance to insecticides (Hinkle et al, 1995, Carlini et al., 1995). The natural flavonoids such as quercetin and gossypol are capable of inhibiting GST (Wood et al., 1990). That is one more fact that can explaine toxic effect of quercetin on the gypsy moth larvae.

Our results show the trend of increas in the amount of GSH in the midgut tissue of the caterpillars which were fed diet suplemented with quercetin (Table 1). Reduced glutathione can also react passively as an antioxidant and can restitute enzymes by reduction of oxidized SH groups (Jocelson, 1962).

It is well known that feeding on certain host plants can alter the susceptibility of the herbivore to insecticides (Berry et al., 1980). The herbivorous insects metabolize and detoxify insecticides using the same enzymes that are involved in the metabolism of ingested plant allelochemicals (Brattsten 1979). Induction of a detoxification and antioxidative enzyme system as a result of feeding on particular host plants can alter to susceptibility to insecticides (Berry et al., 1980).

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Summary

RESPONSE OF THE ANTIOXIDATIVE DEFENCE TO FLAVONOID QUERCETIN IN TWO POPULATIONS OF *LYMANTRIA DISPAR* L.

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The gypsy moth caterpillars used in this experiment were originating from two populations (oak and locust tree forest) which were differently adapted to toxic effects of quercetin supplemented in artificial diet. The responses of 4th instar *Lymantria dispar* L. to artificial diet with quercetin (1.5% w/w) were monitored at the level of antioxidative defence in the midgut tissue: the activities of superoxide dismutase (SOD), catalase (CAT), glutathione-S-transferase (GST), glutathione reductase (GR) and glutathione content (GSH). Regardless of population origin activity of SOD was higher in the caterpillars if fed diet with quercetin than in a control group. In average SOD and CAT activities were higher in the population from locust tree forest than oak forest population. An artificial diet with quercetin led to a decrease of GST activity in both populations. The diet with quercetin did not affect activity of CAT and GR.

CHRYSOLINA FASTUOSA (COLEOPTERA: CHRYSOMELIDAE) AN AGENT FOR BIOLOGICAL CONTROL OR A PEST?

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The species diversity of our planet is one of the most important maintaining resources of life and thus, it represents an integrated element of the sustainable agricultural production. There are innumerable hardly visible living organisms contributing almost invisibly to the welfare of mankind. Regarding the species richness, the Insecta class of the phylum Uniramia (formerly Arthropoda) contains the most species living on the Earth. Insular and selfish human activity called production of goods or free market economy managed to put the life sustaining mechanism of our ecosystem with ill-considered destruction of living beings in danger. The following study on a barely known insect might direct our attention to our common responsibility and interest for a better appreciation of the services of the insects.

Chrysolina fastuosa (Scopoli, 1763) is a chrysomelid beetle with 5-6 mm body length. Head, thorax, elytra are shining greenish golden. It is widely distributed in continental Europe, but of very local occurrence in Britain. In Europe recent sources reported its occurrence in Belgium (Varlez, 1988), Hungary (Víg, 2001), Transylvania (Rozner, 1998) and Bohemia (Rehounek, 2002). In Hungary adult beetles can be found from April to August or September on hemp-nettle (*Galeopsis pubescens*), black horehound (*Ballota nigra*) and surely on other labiatae plants. Since *Ch. fastuosa* can be seen very often in strikingly great number on black horehound, this fact urged us to have a closer look at it.

The aim of this short study was to assess the controlling capacity of *Ch. fastuosa* on *B. nigra* an on roadsides and along walls commonly occurring soft caulescent European plant which can be called minor weed.

Materials and Methods

A with black horehound densely grown area to be found on a roadside in Gödöllő (small university town in the north of Hungary) has been chosen for the observation in the spring of 2003. Forty *B. nigra* plants with adult *Ch. fastuosa* have been randomly selected and labelled. The height, the number of leaves of the plants as well as the number of beetles feeding on their leaves and the caused surface damage have been measured and counted

on 24th May and a week later. The plants have been selected according to their height into three groups (plants to about 20, 30 and 50 cm height) in order to see the possible preference of *Ch. fastuosa*. The progress of damage and the change of the beetles' number were calculated and compared. For considering remarkable differences two-tailed t-test was used (Sváb, 1981).

Results and discussion

Results are presented in Table 1 and 2.

Table 1. Number of *Chrysolina fastuosa* adults on *Ballota nigra*

Date of evaluation	Plants of 20 cm	30 cm	50 cm
0524	1.46[a] (0.660)	4.93[a] (4.714)	4.61[a] (6.158)
0530	1.73[a] (1.555)	3.12[a] (3.638)	10.71[a] (8.789)

Standard deviation is in brackets. Means followed by the same letter within a column are not significantly different at P = 0.05 by two tailed t-test.

Table 2. Surface damage caused by *Chrysolina fastuosa* adults on *Ballota nigra*

Date of evaluation	Plants of 20 cm	30 cm	50 cm
0524	25.08[a] (18.518)	40.71[a] (28.280)	23.57[a] (17.568)
0530	56.09[b] (19.852)	68.47[b] (15.910)	44.00[a] (25.538)

Standard deviation is in brackets. Means followed by the same letter within a column are not significantly different at P = 0.05 by two tailed t-test.

Although, in two height groups regarding the number of individuals feeding on *B. nigra* some progress could have been observed, no significant difference was revealed between the two evaluations (table 1). In contrast to that, results in table 2 show that in two height groups significant differences have been found and also in the third group a remarkable not significant damage progress could be perceived which means that the control of *Ch. fastuosa* on *B. nigra* can be considerable. Hence, theoretically this shining beetle seems to contribute to the control of a minor weed for that reason it is worthy of further research.

However, taking into consideration the growing importance of the production of medicinal plants among which Lamiaceae species are numerous, *Ch. fastuosa* can cause a damage not known yet in more detail.

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Summary

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A poorly known chrysomelid beetle, *Chrysolina fastuosa* (Scopoli, 1763) has been found in strikingly great number on black horehound (*Ballota nigra*), a soft caulescent plant belonging to Lamiaceae family. *B. nigra* can be found from April to August commonly on roadsides, along walls or at the border of gardens and orchards in Hungary and continental Europe, thus it can be called a minor weed. The glistering tiny adults feeding voraciously on its leaves caused apparently important damage. The subsequent investigation focusing on the number of feeding individuals and the loss of plant tissue showed 1-24 beetles a plant and the consumed leave surface amounted 8-94 %. The repeated damage assessment a week later pointed out a significantly unimportant increase in the number of individuals but a significantly considerable, 20-31 % increase of the plant loss. According to the literature *Ch. fastuosa* has been found in association with other labiate plants such as *Galeopsis pubescens*, *Lamium alba* and *Urtica* spp. (Urticaceae) thus regarding its efficiency presented above it could be used augmenting and maintaining its populations as biological control agent of these weeds. However, taking into consideration the growing importance of the production of medicinal plants among which Lamiaceae species are numerous, *Ch. fastuosa* can cause a damage not known yet in more detail.

ACUTE EFFECT OF CADMIUM ON PHOSPHATASE ACTIVITY IN THE MIDGUT OF GYPSY MOTH LARVAE

(Summary)

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The effects of two cadmium concentrations (10 and 30 gCd/g dry food) on larval mass and midgut phosphatase activity (total acid, lysosomal and alkaline) as well as their plasticities were investigated in the 4th instar larvae of the gypsy moth (*Lymantria dispar* L.) under acute three day exposure to cadmium. The analysis was performed on 20 egg masses (5 larvae/egg mass/treatment). It was found that acute exposure to lower cadmium concentration had inhibitory effect only on lysosomal phosphatase. Activity of akaline, total acid phosphatase and larval mass remain at the control value. Cadmium concentration of 30 μgCd/g significantly decreased larval mass, and activity of alkaline and lysosomal phosphatases. Activity of alkaline phosphatase had greater plasticity at 30 than 10 μgCd/g while other traits did not show significant difference in phenotypic plasticity and its variability between the two cadmium concentrations. Acute exposure to both cadmium concentrations increased the variance for larval mass while variability of phosphatase activities were not affected. significant correlations between control group and treatments were not observed while correlations between the environments with different cadmium concentrations were significant only for alkaline phosphatase activity. As midgut homogenates were pulled within each egg mass (full-sib family), the change in a trait variance represents the change in genetic diversity. Additionaly, the absence of significant correlations among environments point to an independent genetic determination of a trait in different environments.

FATE OF IMIDACLOPRID IN SOIL AND PLANT AFTER APPLICATION TO COTTON SEEDS

(Summary)

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The study aimed to investigate the persistence of imidacloprid in soil after seed dressing of cotton and to obtain a complete picture on the mass balance of this compound in soil and cotton plants. The study was carried out as a pot experiment under laboratory conditions using a Gaucho formation containing ^{14}C-labelled imidacloprid. Three treatments of treated cotton seeds were made in sandy loamy soil: fertile seeds grown in autoclaved soil, dead seeds put in fertile soil and fertile seeds grown in fertile soil. Results showed that total ^{14}C recoveries decreased by time from 93.8 – 96.2, 77.1 – 88.4, 53.5 – 62.4 and 60.0 – 64.5% of the applied radioactivity at days 7, 14, 21 and 28 after application, respectively. Extracted ^{14}C from soil decreased in all treatments by time up to three weeks following application and this coincided with fluctuated increase of non-extracted ^{14}C (bound ^{14}C). Bound ^{14}C level was always less in autoclaved soil than in fertile ones. Results revealed also that only 1.8 – 6.8% of the applied ^{14}C was taken up by the plants and fluctuated within the test period. ^{14}C levels were higher in plants grown in autoclaved soil than those in fertile ones. Radioactivity tended to accumulate on the edges of cotton leaves. Most of the radioactivity in the soil extracts was identified as unchanged ^{14}C-imidacloprid. The residues of this compound in soil declined with time with half life periods ranging from 12.6 to 14.3 days at 24 ºC. This decline was due to a rapid degradation process of imidacloprid forming metabolites, which were quickly mineralized or fast bound to the soil matrix since higher concentrations of ^{14}C-metabolites were not detected. The degradation products were changed to $^{14}CO_2$ or might also be associated with the non-extracted residues that were analyzed quantitatively.

POSTER SESSION
WEED SCIENCES & INTEGRATED PEST MANAGEMENT (IPM) GROUP

DETERMINATION OF RESISTANT BIOTYPES OF *AMARANTHUS RETROFLEXUS* L. ON ALS INHIBITORS

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Adaptive ability of weed species during individual development on herbicide action is ontogenetic adaptation, gradual and hardly noticeable. It occurs as the consequence of long lasting action of the particular herbicide during several years lasting application. Weed resistance occurrence has great practical significance in current situation when herbicides are applied in high in almost all countries of the world. Resistance causes inability of previously successful herbicide control of certain weed species, i.e. their lower systematic categories that are more variable and unstable than the species itself. Therefore, mass occurrence of resistance within a weed species can be major limiting factor of further herbicide application. Resistance phenomenon cannot be distinguished by visual assessments until 1-10% of resistant individuals of one population in the field do not occur, i.e. less than 0.1% in laboratory conditions. If only one weed species is spread, it can be considered that there occurred resistance (Konstantinovic, 1999).

Literature

In 1970 weed herbicide resistance was for the first time established in Washington forest nursery, where *Senecio vulgaris* resistance on triazines was determined (Holt, 1992, cit. Konstantinovic and Meseldzija, 2002). Following several repeated treatments found some weeds such as *Solanum nigrum, Amaranthus retroflexus* and *Chenopodium album*, with anatomic modifications on the membrane level became unsusceptible to this herbicide type. Evolution of resistant weed species was caused by frequent use of the herbicides belonging to the herbicide group with the same mechanism action in over five years period.

Crossed resistance was also determined in cases in which a weed biotype is resistant to two or more herbicides due to the presence of one resistance mechanism, as well as multiple resistance i.e. occurrence of resistant plants to herbicides that have two or more resistance mechanisms. All of them make choice of alternative herbicide as mean of control in the case of resistance more complicated. Therefore, rotation of various herbicides with

various mechanisms of action is necessary. *Lolium rigidum* in Australia and *Alopecurus myosuroides* in Great Britain are examples of the occurrence of crossed, i.e. multiple resistance (Le Baron, 1991).
Up to day, HRAC group (Herbicide Resistance Action Committee), founded in 1989 by industry with support of Federation for global crop protection registered 272 resistant weed species on over 210.000 fields, of which even 79 species developed resistance on ALS inhibitor herbicides (HRAC, 2003). In 1988 herbicide resistance in our country was for the first time determined in *Amaranthus retroflexus* L. (Janjic et al., 1994). In 1991, as the result of more years lasting use of triazine herbicides on Yugoslav railroad tracks, Arsenovic et al. also determined resistance in various weed species, such as *Amaranthus retroflexus* L., *Convolvulus arvensis* L., *Sorghum halepense* (L.) Beauv., *Cynodon dactylon* (L.). Pers etc. In 2001, in studies performed by Konstantinovic and Meseldzija in certain localities of Vojvodina, resistance of biotypes of *Amaranthus retroflexus* and *Setaria viridis* was determined. This was confirmed by Herbicide Resistance Action Committee (HRAC, 2003).
We considered studies of resistance occurrence of the weed species *Amarantus retroflexus* L. on ALS inhibitors, group of herbicides that include sulfonilureas and imidazolinones, of which imazethapyr was applied the most frequently in our country last years, of great importance. Primary site of ALS inhibitor action is enzime acetolactate synthaze that has key role in biosyntesis of amino acid leucine, isoleucine and valine in weed plants. Inhibition of these amino acids synthesis causes reduction of protein quantity in younger parts of the plant, and reduced protein synthesis inevitably leads to inhibition of cell division. Species *Amaranthus retroflexus* L. is the first weed in Israel that developed resistance to ALS inhibitors (Sibony & Rubin, 1996). Studies of weed species *Echinochloa crus-g*alli L. resistance on ALS inhibitors are the first studies of weed resistance occurrence toward herbicides with this action mechanism in our country. Up to now, resistance of the species *Amaranthus* retroflexus L. has been determined in Israel, Canada and four USA states (HRAC, 2003).

Materials and Methods

Seeds were collected from the different sites of locality Becej, which had a long history of imidazolinone and sulfonilurea herbicides use (ten years backwards). A susceptible population collected from an area where no herbicides had been used was used as a reference population. Imazethapyr was used since it was one of the most frequently applied ALS inhibitors in the localities studied.
The most important individual factor for the initial determination of resistance, is the level of non-susceptibility in the field. Consequently, we

have used a method of visual assessment of imazethapyr efficiency to detect possible resistance. There are several factors that can indicate possibility of resistance occurrence in field, such as: level of control of other susceptible species, presence of live plants alongside dead ones, past experiences, i.e. previously successful control by the same treatment, herbicide history, i.e. repetition of the same herbicide treatment, or herbicide with the same mode of action, resistance occurrence in the region, harvest, cultural history, i.e. monoculture and minimum tillage (Moss, 1995).

Studies were made on whole plants (Thurwachter, 1998) and Petri dishes bioassays (Clay & Underwood, 1990). Assays were performed in four replications and plants were treated with various doses of imazethapyr, representing. 40, 80, 100, 150 and 200 g a.i. ha^{-1}.

In whole plant studies, plants were grown in controlled conditions in pots from seed which was suspected to be imazethapyr resistant. There were ten seeds per plots and the trial was set on chernozem with 3.5% of humus in four replications, and assessments were done 50 days after treatment (pre emrgence herbicide application). In whole plant studies, efficacy was evaluated by measuring height steam, as well as by counting emerged plants and assessing their vigour.

In the Petri dish assays, twenty seeds per dish were spread evenly over the paper and 5ml of imazethapyr solution added to saturate, but not flood, the filter paper (pre emergence herbicide application). There were four replications of each treatment. Dishes were kept in termostat on 22^0C. Germination and seedling condition were recorded at intervals up to 25 days from the start, with visual assessment of number of healthy and damaged seedlings in each dish. In Petri dishes bioassays, the lengths of epicotyls and hypocotyls of shoots were measured.

Results

In Figure 1 mean epicotyls lengths of the *Amaranthus retroflexus* samples from Becej locality are presented, table T-7, T-26 and T-72, as well as of the sample from non-agricultural land that served as standard.

In Figure 2 mean hypocotyls length values of *Amaranthus retroflexus* L. samples from locality Becej are presented, table T-7, T-26 and T-72 and susceptible standard.

Figure 1. Mean epicotyls lengths of the species *A. retroflexus* L. from various localities in range of imazethapyr rates

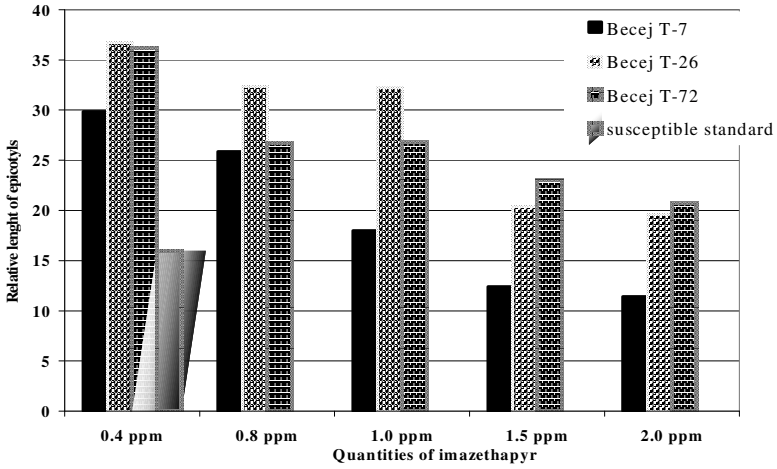
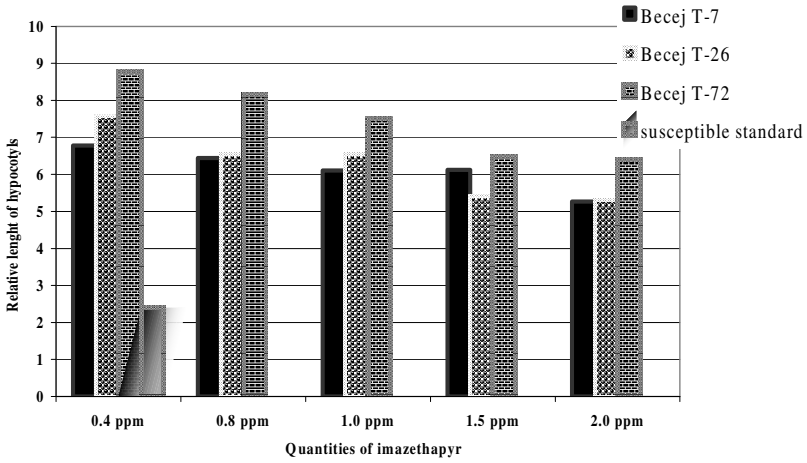
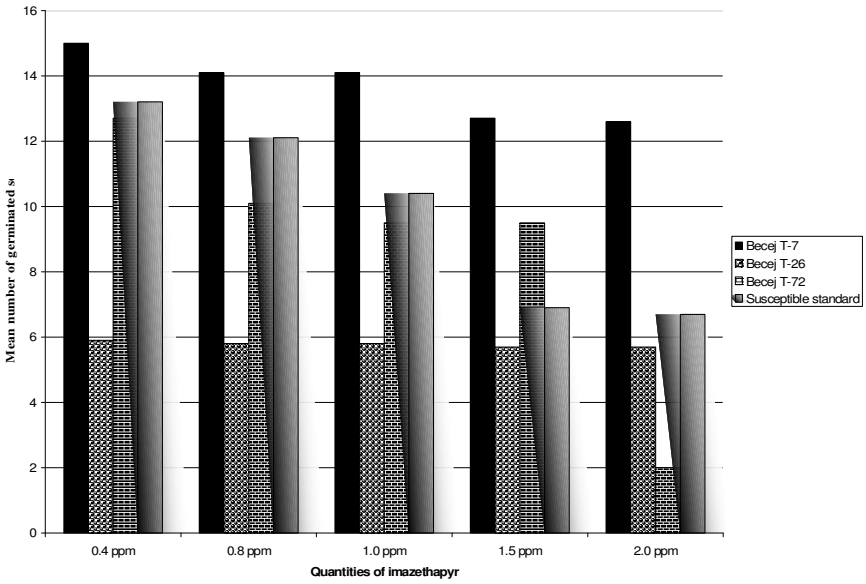


Figure 2. Mean hypocotyls lengths of the species *A. retro*flexus L. from various localities at range of imazethapyr rates



During resistance studies in Petri dish assays, seed germination of the species *Amaranthus retroflexus* L. in different imazethapyr quantities was also measured (Figure 3).

Figure 3. Average number of germinated seeds of the species *A. retroflexus* L. at different herbicide imazethapyr quantities



In whole plant studies, 50 days after emergence stem height was measured, and calculated mean values in range of imazethapyr rates are given in Table 1.

Table 1. Mean stem height of *A. retroflexus* L. at different imazethapyr quantities

Imazethapyr quantity (g ha^{-1})	Mean stem height of A. retroflexus L. (mm)			
	Becej T-7	Becej T-26	Becej T-72	Susceptible standard
40	8,36	8,75	8,10	5,5
80	7,79	7,58	7,67	2,75
100	7,47	6,70	7,33	0
150	6,05	6,25	6,95	0
200	6,01	6,25	6,56	0

Discussion

At Petri dish assays 20 seeds in four replications were germinated in thermostat at temperature of 22 ^{0}C. Three days upon setting of the assay, 65% of germinated seeds were recorded. On the fifth day maximal seed germination was recorded. It was 60% for seeds from locality Becej T-7,

30% for seeds from locality Becej T-26, 50% for seeds from locality Becej T-72 and from non-agricultural land it was 35%.

Ten days upon setting of the assay the best seed germination was determined for samples from localities Becej T-72 and Becej T-7. Later on, in these samples were measured significantly lower values for epicotyls and hypocotyls lengths as regards to the first locality. Seed from locality Becej T-72 had the highest values of the studied parameters. The lowest germination was recorded for seeds from locality Becej T-26, which was lower even from one recorded for seeds collected from non-agricultural land.

During second week, gradual decay arose on shoots of samples from non-agricultural land at higher imazethapyr quantities. Shoots epycotils and hypocotyls lengths from localities Becej T-7, 26 and 72 were significantly lower at imazethapyr quantities of 1.5 and 2.0 ppm. This was especially typical of samples from locality Becej T-7, where no shoot decay occurred.

Based upon values for the mean epicotyls and hypocotyls lengths, from *Figures 1*. and *2*. it is obvious that samples from non-agricultural land and from locality Becej T-7 showed the highest susceptibility at imazethapyr quantity of 2.0 ppm. This was also the case with imazethapyr quantity of 1.0 ppm.

At samples from all three studied localities (Becej T-7, Becej T-26 and Becej T-72), increase of imazethapyr quantity led to gradual decrease of epicotyls and hypocotyls relative length, and higher quantities above 0.4 ppm of imazethapyr led to decay of shoots from non-agricultural land.

In whole plant studies, a month after shooting up of *A. retroflexus* L, the first occurrence of chlorosis at higher imazetaphyr quantities on samples from non-agricultural land and locality Becej T-7 was determined. From *Table 1*. it is obvious that samples from non-agricultural land and locality Becej T-7 had the highest susceptibility at imazethapyr quantity of 100 and 200 g ha^{-1}. Increase of imazethapyr quantity led to gradual reduction of mean stem height values of samples from all studied localities, excluding samples from non-agricultural land. Seed of *A. retroflexus* L. from all localities did not germinate at imazethapyr quantities of 100, 150 and 200 g. a.i. ha^{-1}

According to the obtained results, samples from locality Becej T-72 showed the highest resistance, even at imazethapyr quantities of 150 and 200 g. a.i. ha^{-1}.

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Summary

DETERMINATION OF RESISTANT BIOTYPES OF *AMARANTHUS RETROFLEXUS* L. ON ALS INHIBITORS

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The aim of the study was determination of resistance occurrence of the species *Amaranthus retroflexus* L. on ALS inhibitors. Weed resistance toward herbicides represents phenomenon of these plant species adaptation toward changed environmental conditions and occurs as the consequence of use of herbicides with the same mode of action during several years lasting period. Studies with the aim of resistance determination were performed during 2002, and material for the studies was collected from various sites at Becej locality (Vojvodina). Long lasting use of the herbicides belonging to the group of ALS inhibitors, which are very successfully applied for control of dicotyledonous and monocotyledonous weeds in certain areas lead to occurrence of resistance of the weed species *Amaranthus retroflexus* L. Results of the study obtained by biological assays, field experiments and Petri dish assays confirmed presence of resistant biotype *Amaranthus retroflexus* L.

ALLELOPATHY OF SOME IMPORTANT PERENNIAL WEEDS

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Harmful effect of the neighbouring plants on each other is called interference, which consist of competition and allelopathy. Allelopathy is considered as one of the stress factors, which influence the development of an individual plant, a species or plant community. It plays an important role in plant succession and is a well known phenomenon in natural ecosystems. In agriculture, it has been frequently studied in crop monocultures. The two fundamental approaches to the use of allelochemicals for weed management are: 1. as a herbicide or a lead for a synthetic herbicide and 2. use of allelopathic plants in plant production (Kazinczi 1999, Béres 2000, Duke et al. 2002).

Literature

Perennials cause serious problems in Hungary. *Asclepias syriaca* L. originated from North-America is an adventive weed in Hungary. According to the first Hungarian Weed Survey (1947-1953), it did not exist in Hungary, while on the basis of the IV. National Weed Survey (1996-1997) it occupied the 76th positions (Tóth and Spilák 1998). *Cirsium arvense* (L.) Scop., a native plant of Europe is a weed of 27 crops in 37 countries (Holm et al., 1977, Moore 1975). *Convolvulus arvensis* L. is one of the most troublesome weed in Europe, western Asia, Canada and the United States, and it is a special problem in several crops grown widely in the temperate region (Holm et al., 1977). On the basis of the IV National Weed Survey *C. arvense* is the 5th, while *C. arvensis* is the 6th most important weed in Hungary (Hunyadi et al., 2000). Besides generative reproduction by seeds, their vegetative propagation is also very important and occurs mainly by rootstocks, containing adventitious buds. Rapid distribution of these species in not only due to their considerable reproductive and competitive ability (Evetts and Burnside, 1973, Moore, 1975, Bhownik and Bandeen 1976, Holm et al., 1977, Lehoczky, 1988, Varga, 1998, Hunyadi and Kazinczi 1992, Lehoczky, 2000, Lehoczky et al., 2003), but also due to their allelopathic properties (Bendall, 1975, Wilson, 1981, Kovács et al., 1988, Béres and Csorba, 1992, Solymosi and Nagy, 1999, Béres and Kazinczi, 2000, Kazinczi et al., 1999, 2001).

The aim of our examination was to study allelopathic effect of *A. syriaca*, *C. arvense* and *C. arvensis* in bioassay and pot experiments.

Materials and Methods

Laboratory germination (bioassay)
Fresh rootstocks and shoots of *A. syriaca* and *C. arvense* were collected at the beginning of flowering in Vecse, Somogy county (Hungary) in July 2003. The roots and shoots were cut into small pieces in a grinder. After grinding 25, 12.5, 5 and 2.5 g fresh biomass was stirred into 100 ml distilled water and left for a day. Then the mixtures were filtered through filter paper (MN 640w) and were denoted as a stock solution, 2x, 5x and 10x dilutions. Double filter paper was kept in each Petri dish, thereafter 8 ml leachate was added per Petri dish. On the top of filter paper 100 seeds of cucumber 'Delicatesse' were placed to germinate at 22°C in incubator in four replicates. Germination percentage and the length of radicle were recorded after 48 hours. Seed germinated in distilled water served as control.

Pot experiments
Fresh water extracts was made with 25 g fresh *C. arvense* shoots/100 ml distilled water. Water extract was used for irrigation of the pots as needed. Pots were filled with the soil mixture of sand (pH: 6.96, humus: 0.27%) + peat (pH: 6.78, humus: 9.98%) in a ratio of 1:1 and sown with four seeds of wheat 'Mv-23', corn 'Mv-NK 333', and sunflower 'Barbara' in four replicates.
In another experiment 0.8 kg dried shoots of *C. arvensis* was mixed in 10 kg soil mixture and kept moist for three months. After three months of decomposition, pots were filled with the soil mixture containing shoot residues and sown with four seeds each of three test plants spp. i.e. barley, mustard and bean.
Fresh weight of the test plants was recorded 37 days after sowing (DAS).

Results

Laboratory germination (bioassay)
It has been seemed that inhibition greatly depended on the concentration of the extracts. At higher concentration was a stronger inhibitory effect, due not only to its direct toxic effect but due to increased osmotic potential as well. Radicle length of cucumber was retarded to a greater extent than germination of that. Strongest inhibitory effect was observed in case of *A. syriaca* root water extract, which at the highest concentration reduced radicle length of cucumber by nearly 100% (Table 1). In our previous experiments similar strong inhibitory effect on the germination of sunflower

and *Amaranthus retroflexus* L. was observed with *A. syriaca* shoot and root water extracts, respectively. On the contrary soil incorporated root residues of milkweed stimulated the growth of all test species (Kazinczi et al., 1999, Béres et al., 2001). Our results confirmed to those of Béres et al. (2001), that water extracts of *C. arvense* did not influence the germination of cucumber, however alcoholic ones significantly reduced germination of test plants.

Table 1. The effect of water extracts on the radicle length and germination of cucumber

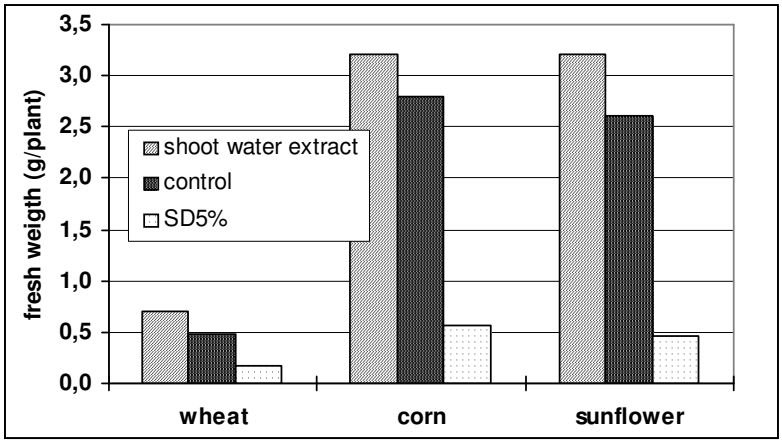
		radicle length (mm)		germ %	
***C. arvense* shoot**	***S****	30	$SD_{5\%}$=12	86	$SD_{5\%}$=17.6
	2x	39		79	
	5x	62		81	
	10x	59		90	
	C	64		81	
***C. arvense* root**	***S****	45	$SD_{5\%}$=16.8	82	$SD_{5\%}$=15.2
	2x	67		91	
	5x	60		92	
	10x	78		90	
	C	64		81	
***A. syriaca* leaf**	***S****	27	$SD_{5\%}$=7.6	86	$SD_{5\%}$=7.5
	2x	55		84	
	5x	64		85	
	10x	67		87	
	C	62		91	
***A. syriaca* stem**	***S****	11	$SD_{5\%}$=12.3	81	$SD_{5\%}$=7.3
	2x	53		90	
	5x	61		91	
	10x	61		88	
	C	62		91	
***A. syriaca* root**	***S****	1	$SD_{5\%}$=5.3	69	$SD_{5\%}$=6.3
	2x	5		98	
	5x	25		96	
	10x	40		92	
	C	41		98	

*S, stock solution (25g fresh plant part/100 ml distilled water); 2x, two fold dilution; 5x, five fold dilution; 10x, ten fold dilution; C, control

Pot experiments

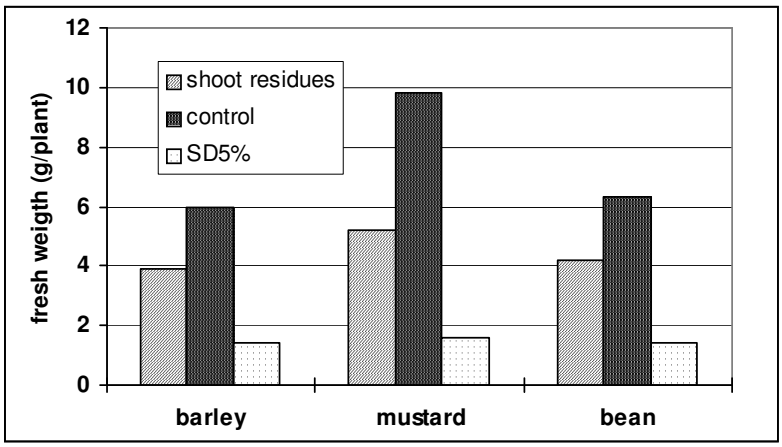
Fresh shoot water extracts of *C. arvense* used for irrigation significantly promoted the growth of sunflower and wheat, while had no significant effect on corn (Figure 1). Similar, promoting effect was observed with *Abutilon theophrasti* Medic. water extract (Kazinczi et al., 2001, Béres et al., 2001) and leaf leachates of several cereal weeds (Kazinczi et al., 1997).

Figure 1. The effect of water extracts of *C. arvense* shoots on the development of the test plants in pot experiments



Shoot residues of *C. arvensis* incorporated into the soil significantly reduced the fresh weight of all test species (Figure 2). The inhibition followed the order: mustard › barley › bean.

Figure 2. The effect of shoot residues of *C. arvensis* on the development of the test plants in pot experiments



There are a lot of papers about inhibitory effect of plant residues, although opposite effect had been observed in some cases, when phytotoxins present in fresh plant parts gradually decompose due to the microbiological degradation in the soil (Kazinczi et al., 1991, Kazinczi et al., 1999, Béres et al., 2002).

Discussion

It has been seemed that allelopathic effect depends on a lot of factors, i.e. donor and recipient species (varieties), their phenological stages, different plant parts, concentration and method of preparing of the extracts, etc. Latest results indicated that environmental factors (i.e. nutrient and water supply) also influence allelopathic potential (Dávid and Radócz 2002). Nevertheless future investigations are necessary in order to identify allelochemicals from crops and weeds, and to search their application in weed management under field conditions. In allelopathic research, elaboration of a uniform investigation method would be very important in order to better comparison of scientific results.

Acknowledgement

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Summary

ALLELOPATHY OF SOME IMPORTANT PERENNIAL WEEDS

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Laboratory (bioassay) and pot experiments were carried to study allelopathic effect of some important perennial weeds, i.e. *Cirsium arvense, Convolvulus arvensis* and *Asclepias syriaca*. In germination tests it has been seemed that inhibition greatly depended on the concentration of the extracts. At higher concentration was a stronger inhibitory effect, due not only to its direct toxic effect but due to increased osmotic potential as well. Radicle length of cucumber was retarded to a greater extent than that of germination %. Strongest inhibitory effect was observed in case of *A. syriaca* root water extract, which at the highest concentration reduced radicle length of cucumber by nearly 100%. Fresh shoot water extracts of *C. arvense* used for irrigation significantly promoted the growth of sunflower and wheat, while had no significant effect on corn. Shoot residues of *C. arvensis* incorporated into the soil significantly reduced the fresh weight of all test species. The inhibition followed the order: mustard › barley › bean.

THE OCCURRENCE OF SPECIES *AMBROSIA ARTEMISIIFOLIA* L. ON THE TERRITORY OF BIHOR COUNTY

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The presence and expansion of *Ambrosia artemisiifolia* L. in the Western Plain of Romania is a real fact, confirmed by the results presented in this short paper.

Central European countries such as: Germany, Czechia, Hungary and countries from the Western part of the continent such as: France, Portugal, Spain and so on, have paid a growing attention to the very strong allergic phenomena, caused by this very dangerous plant, that is included in the group of quarantine weeds.

Despite this fact, countries from Eastern Europe, like Romania, have ignored this phenomenon by not taking the appropriate measures of stopping the spreading of this weed.

After 1991, a crucial year for the Romanian agriculture (because of the new agricultural reform), significantly large ploughlands have remained uncultivated, fact which led to favourable conditions for the development of the weeds. One of the weeds, that have found suitable conditions to get rooted on these surfaces, is *Ambrosia artemisiifolia* L.

Nowadays, we can frequently find it on the two sides of the roads and railways, on the plots, with agricultural crops, found in the immediate proximity of these; at present it is signalled both in the steppe area and forest steppe, reaching the oak tree floor. Its occurrence seems also possible in other geographic areas, even if these individuals have grown out of seeds brought from other areas; here, they can't produce fruits due to the climate, and thus, they have no chance to form populations.

There have been written and published many scientific papers on *Ambrosia artemisiifolia* L. and *A. elatior* L. Researchers from different periods of time have dedicated ample studies to the observation and research of this plant species. Among these, we only mention a few: Palliser, 1863, Hegi, 1906, Jávorka, 1910, Crocker, 1916, Boros, 1924, Moesz, 1926, Kovacevic, 1948, Lavaree, 1955, Priszter, 1957, Wagner and Beals, 1958, Bassett and Teresmae, 1962, Gebben, 1965, Dickerson, 1968, Erdős, 1971, Béres, 1979, 1981, Béres and Hunyadi, 1980, 1991, Claupein, 1994, Rybnicek and Jager, 2001, Kazinczi et al., 2002, Dechamp and Meon, 2002, and many others.

Thus, the weed has been signalled for the first time on the present territory of Romania, in the year 1908 (Hegi), in Banat area (during that period, this area was under the Austro-Hungarian administration); then it has been signalled at Sighet (Topa and Boscaiu, 1965), in Moldavia at Huşi and Bârlad (Mititelu, 1970) and in Muntenia at Ploiesti (Negrean, 1971), cited by Hodisan, in 2003. In 1968, Sanda et al. have written on the spreading of this species in the agricultural crops or other places.

Materials and Methods

During the travels that we have undertaken in order to localize these species, so as to bound the coverage area in the Bihor county, we have been unpleasantly surprised to notice that, due to the carelessness of the people, coroborated with the disinterest of the competent authorities, *Ambrosia artemisiifolia* L. has pervaded the agricultural cultures, such as: maize, sunflower, sugarbeet, tobacco, as well as the park and entertainment touristical areas.
During the travels undertaken on the previously mentioned routes, the main ecological areas of the county have been covered, determining the presence and spreading extent of this species. Some samples have been collected for the future determinations regarding the viability of the polen and seeds issued from the four areas.

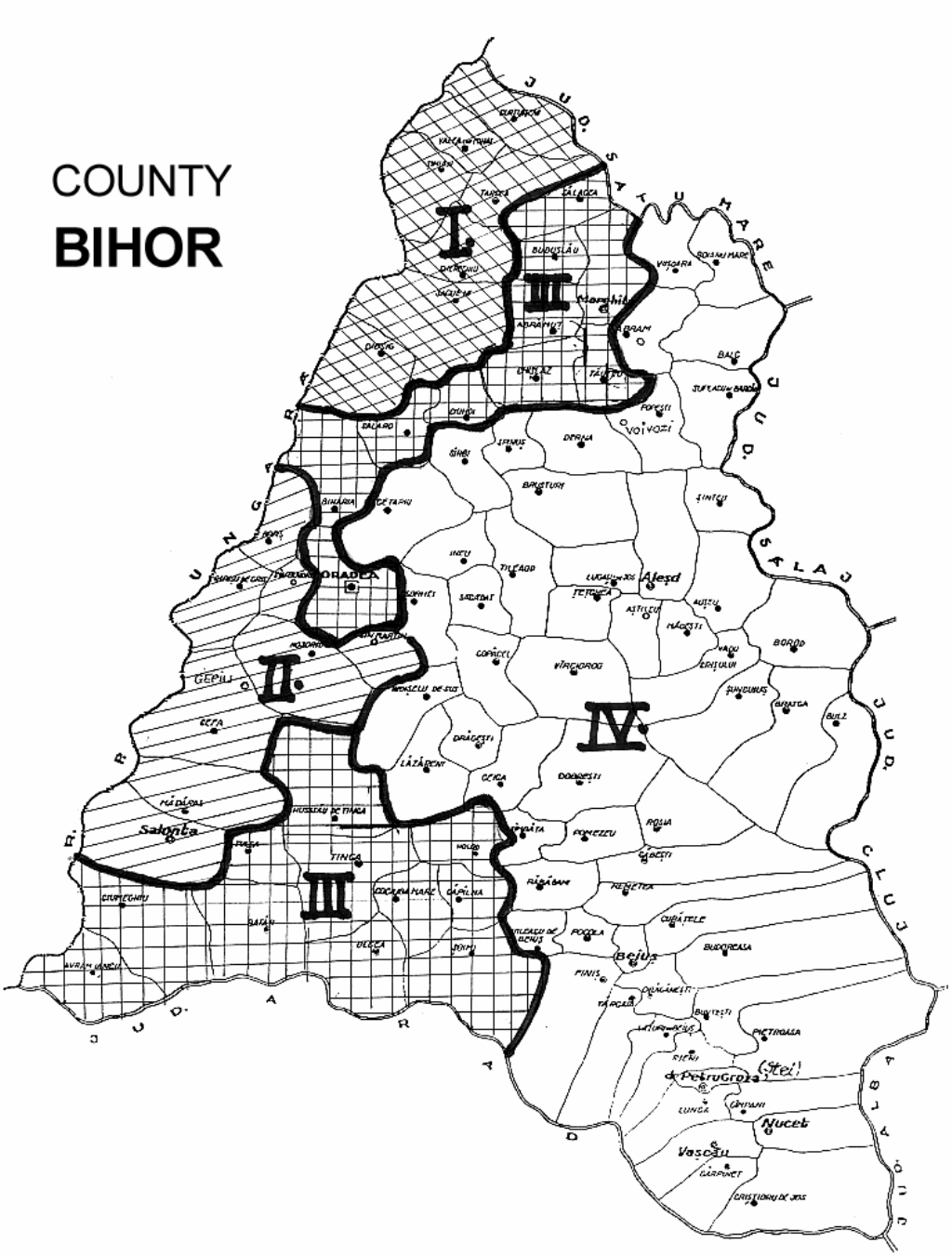
Results

In order to support some concrete control measures against the expansion of Ambrosia artemisiifolia L., known in Romania under the folk name of "Pusta Flower", we will present the limits of the coverage areas according to the occurrence and spreading extent, on the map of the Bihor county.

- Area I, located in the North-Western part of the Bihor county, along the Ier Valley, is characterized by the existance in this area of some large surfaces which usually surpass more hectares, where *A. artemisiifolia* L. has found the most favourable living conditions; due to the transport of cereals or other raw materials that come from this area, new territories are fed with mature seeds ready to germinate where they find favourable conditions (Valea lui Mihai, Curtuişeni, Tarcea,Otomani, Silindru, Cheşereu, Şimian, Cherechiu, Săcuieni, Diosig).
- Area II, located in the Western part of the Bihor county, is characterized by the presence of numerous populations of *A. artemisiifolia* L., which contain individuals grouped in cut-off trenches large of several up to tens of square metres. The area is of major interest because the species has found here the pedoclimatic conditions favourable to reproduction

(Borş, Sântandrei, Girişu de Criş, Sântandrei, Nojorid, Gepiu, Cefa, Sânmartin, Mădăras, Salonta).

Figure 1. Map of Bihor county indicated the infected areas by *A. artemisiifolia*



- Area III, which forms a cord on the North-Southern axis of the Bihor county, in the immediate proximity of area I and II, is characterized by the fact that within this area there have been signalled individuals of *A. artemisiifolia* L. that live either isolated or in small groups of several individuals. These individuals don't succeed in reproducing themselves, because in this area the seeds can't reach adulthood, but it is an area continuously fed with mature seeds issued from areas I and II with which it is in close proximity. Here, the plant is also considered to be dangerous, because it flourishes and produces polen in significant quantities (Salacea, Buduslău, Marghita, Abrămuț, Tăuteu, Chişlaz, Ciuhoi, Sălard, Biharia, Husasău de Tinca, Tinca, Tulca, Ciumeghiu, Avram Iancu, Batăr, Olcea, Şoimi, Cociuba Mare Căpâlna).
- Area IV, which is delimitted from the beech floor upwards, is an area where the individuals of *A. artemisiifolia* L. species can be found very rarely; here, even if this species flourishes, the polen that has been produced, is in insignificant quantities to produce allergies.

Conclusions

In conclusion, *A. artemisiifolia* L. species have also pervaded the Romanian territory, where as it has been shown above, they have found the most favourable vegetating conditions, thus becoming a real threat for our fellows' health. For this reason, it is necessary as the Romanian authorities to officially declare the existance of this plant on the Romanian territory and through the elaboration of legislative documents to impose measures fighting against the Ambrosia artemisiifolia L. species.
It has been highlighted the necessity of continuing the research regarding the viability of the polen and seeds issued from different ecological areas as well as the necessity of establishing the most appropriate control measures as regards the presence and expansion of the species.

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Summary

THE OCCURRENCE OF SPECIES *AMBROSIA ARTEMISIIFOLIA* L. ON THE TERRITORY OF BIHOR COUNTY

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The purpose of this research was to investigate the presence and the spreading area of common ragweed *Ambrosia artemisiifolia* L. in Bihor county, Romania. The investigation was made in the field and the samples were analized in the laboratory confirmed the presence and the expansion of the spreading area of this dangerous weed in the western part of Romania**.** The ecological areas with different densities were also presented as regards the presence of the weed, regarding for the establishment of some control measures against this weed species.

CHANGES IN NUTRIENT CONTENT OF COMMON MILKWEED (*ASCLEPIAS SYRIACA* L.)

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Introduction

The formerly cultivated *Asclepias syriaca* L. has become a weed plant again and was first recognised by forest managers in Hungary almost fifty years ago when fallow lands were to utilise by forests. Its chemical weed control has been introduced only in forest areas.

Common milkweed is native in North America. Its vegetative reproduction is very important. According to one observation, one plant can grow 54 propagation roots and 94 young plants on them during a four-year period. It causes yield loss in maize and in soybean, 5-10% and 12-19%, respectively (Hunyadi 1988). Common milkweed as a weed plant causes substantial damage in crop production.

There is a wide variety of its name in the Hungarian language (krepin, krepinfű, selyemkrepin, selyemfű, selyemvirág, vaddohány, pulykavirág, papagájvirág, mézgyapot). *A. syriaca* belongs to the subclass *Asteridae*, order *Gentianales*, family *Asclepiadaceae*, subfamily *Asclepiadoideae* and genus *Asclepias*. Milky weed can be utilised in many ways.

It came to Europe in 1629 and was tried to use as an industrial crop in France and in Germany, in the 18th -19th centuries. It brought high hopes (silk, paper, fibre, caoutchouc, oil, raw material production), which then failed: "its beauty misses its usefulness" (Hollendonner 1915).

Its main area of distribution is Canada, USA, Iraq, Hungary, France, Switzerland, Germany, Poland, Caucasus, the Baltic Sea and its region. Common milkweed is an adventive, serious weed of Hungary. It can be found mainly in the Southern part of Hungary on sandy soils (Ujvárosi, 1973).

The rapid spread of this weed was observed from 1980. During the first National Weed Survey (1947-1953) it did not appear on the list of species. According to the result of the second National Weed Survey it came out as the 218th, while the third (1987-1988) and fourth (1996-1997) National Weed Survey put it on the 113th and 76th places, respectively, in the order of importance of weed cover percentage. According to the National Weed Survey results in 1989 more than 16.000 ha of arable lands were infected and it also appeared in forests and orchards. The most severely infected

counties are: Bács-Kiskun, Tolna, Jász-Nagykun Szolnok, Somogy and Pest (Varga, 1998).
It causes substantial damages; the weed kills the cultivated plants on the weed-grown fields resulting in considerable yield loss. It grows on river flats too, and kills native plants. Allelopathy may play an important role in its distribution (Kazinczi et al., 1999, Béres and Kazinczi, 2000, Béres et al., 2001).
Its sudden distribution in the past few years can be explained by dry weather conditions in the past ten years, the increase in frost-free days, the lack of stubble ploughing, soil disturbing, minimum tillage, the killing of competitor plants and infrequent selective weed control (Kőrösmezei, 2000).
Its special plant physiological features contribute to the difficulties in its weed control. It can safely described as dangerous. Its vegetative and generative reproduction is successful. It is a highly competitive plant with both cultivated and weed plants. It tolerates herbicides, regenerates rapidly. Its distribution in arable lands, rural or uncultivated areas, pastures, acacia-forests and in fallow grounds keeps increasing.
Contrary to the fact that milky weed is attacked by pests and pathogens, the decrease in its plant population density cannot be expected. Only the regular, planned and integrated weed control of the perennial weeds, including *Asclepias syriaca*, can be successful.
It is crucial for the effective and economical perennial weed control to apply precision and site-specific weed control on weed patches, especially spraying precision herbicides (Reisinger, 1997, 2000; Reisinger et al., 2002b).
The GPS method can make it possible to determine the exact location of these perennial patches and it may play an important role in perennial control in the future (Reisinger et al., 2002a, 2002c). According to American experiences the elimination of *Asclepias* colonies, cutting its root system and chemical weed control together proved to be effective.
Perennials uptake considerable amounts of nutrients from the soil (Lehoczky, 1988, 1994, 2000; Lukács et al., 1998; Radics, 2002).
In our experiment we followed the nutrient uptake and its changing in *Asclepias syriaca* L.

Materials and Methods

A. syriaca plants were collected from the fields round of village Vése in Hungary from May to November in 2002. The main characteristics of this soil are below (Table 1).

Table 1. Characteristics of the experimental soil

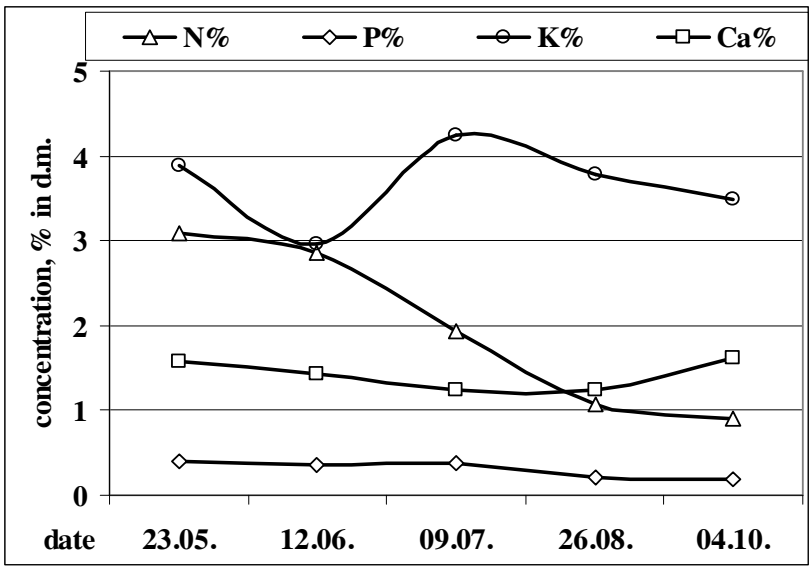
Humus:	2.33 %
AL-P_2O_5	320 mg/kg
AL-K_2O	364 mg/kg
PH $_{(H2O)}$	6,35

Plant samples were collected from the fields (shoots, and roots from the upper 50 cm soil layer) from May to November. We examined nutrition element concentration (N, P, K, Ca % in dry matter) of the samples. We measured the fresh weight of plants and after 40 C° drying the dry mass weight too. Nitrogen concentration was determined by Kjeldahl method, phosphorus concentration by spectrophotometer, potassium and calcium concentration by flame photometer.

Results and Discussion

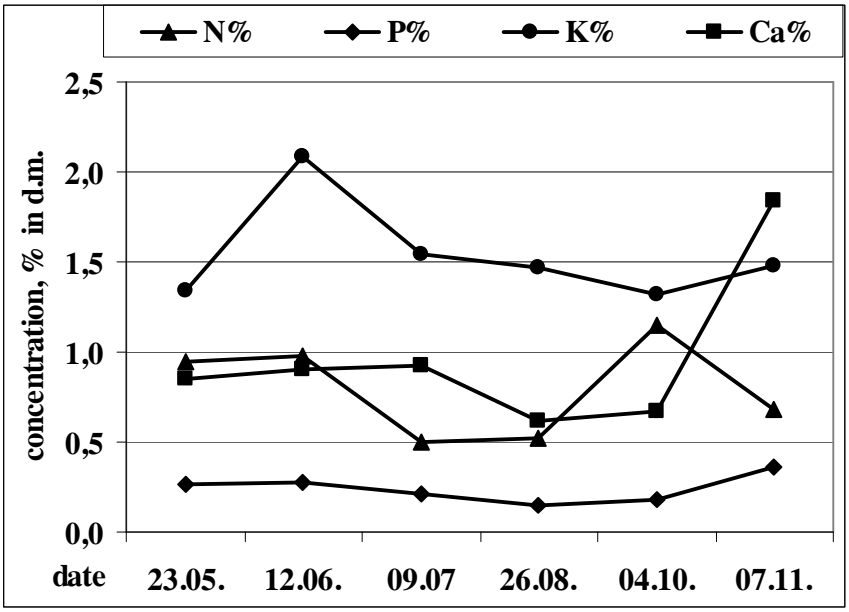
Nitrogen concentration of shoots altered between 0.9-3.1%. Nitrogen concentration of the shoots showed a continuously decreasing trend during the examination period (Figure 1). The nitrogen concentration of the reproductive roots ranged between 0.5-1.1%. The smallest N content was found in July and August, which can be explained by the intensive growth of propagation roots (Figure 2).

Figure 1. Nutrient concentrations in shoots of *Asclepias syriaca* L.



Changes of nutrients concentration of shoots and roots would connect with physiological processes of the plants (Lehoczky, 2000).
Phosphorus concentration of the plants varied within the smallest range between 0.15-0.4%. Phosphorus concentration of the shoots 0.18-0.40 was higher than that of the roots 0.15-0.3% until October.
Phosphorus content in the shoots reached its highest quantity just before flowering in May, and also during the intensive flowering stage in June and July (Figure 1).
Among the elements included in the study Potassium content was found to be the highest both in shoots and in propagation roots (Figure 1, Figure 2). Potassium content in shoots 2.9-4.2% was much higher than in propagation roots 1.3-2.1%. The initial decline in Potassium content (May, June) can be explained by the intensive shoot growth and by its dilution in the biomass synthesised in a large amount. Following the intensive Potassium uptake the Potassium content in the shoots increased remarkably, which can be explained by the intensive flowering and metabolic processes.
The Potassium content in the roots was initially increasing and its quantity measured in May remained on a steady peak level. This finding can be explained by the reserve nutrient accumulation in roots and by the intensive carbohydrate metabolic processes.

Figure 2. Nutrient concentrations in roots of *Asclepias syriaca* L.
metabolic processes.



Calcium concentration was found to be higher 1.2-1.6% at each of the growth stages measured than that in the propagation roots 0.6-0.9%. By the end of the growing season the Calcium concentration in shoots was increased with the simultaneous decline of Calcium concentration in the roots.

Conclusions

Asclepias syriaca can be described as a perennial weed plant with high nutrient demand. Its nutrient (N, P, K) uptake remains intensive throughout the whole growing season. According to our examination *Asclepias syriaca* L. can uptake potassium in a great quantity. This nutrient can be found in high concentration both in its shoots and propagation roots, between 1,3-4,2 %.

During the initial vegetative growing period it takes up nitrogen in large quantities thus resulting in high nitrogen concentration in the shoots (3,1%). The difference between potassium content in shoots and in propagation roots is smaller than regarding the other elements.

Calcium concentration in shoots remains steady and shows an increasing trend towards the end of the growing season.

We established that intensive nutrient uptake has an important roll in considerable competitive capacity of *Asclepias syriaca* especially in competition for nutrients.

Acknowledgement

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HYBRID-SPECIFIC WEED CONTROL IN MAIZE PRODUCTION

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The cereals play a definite role in Hungarian crop production, especially the production of wheat and corn. The national corn production went through a significant transformation during the past decades. The inbred hybrids that have been introduced very fast at the beginning of the 1960's ensured an adequate genetic basis for the wide use of industry-originated inputs. From the '60's the use of fertilizers dynamically improved, the chemical herbicides spread widely, modern machines, appliances were used. In the production-technology of corn the role of agro technological factors have been defined by Győrffy (1976) in the following: fertilizing 27%, hybrid 26%, cultivation 24%, plant density 20%, tillage 3%.

In the 1980's the national corn production's world standard was well characterized by the about 6,0 t ha^{-1} national yield and the fact that in that decade the fluctuation of national yield was really moderate (10-20%). From the beginning of the 1990's –because of the widely known financial and economical difficulties- the yield of corn –depending on the year- decreased by 0,5-2,0 t ha^{-1}, but because of the use of low level industrial inputs the fluctuation significantly increased (30-50% in the case of the national yield).

One of the important element of corn production is the use of herbicides. The modern weed control should be based on the integrated principles (Berzsenyi, 2000, Széll et al., 1985, Chui et al., 1997). Lately, in the national corn production the significance of early and normal post treatment considerably improved, where the appearance and composition of weed is known. The effect and efficiency of postemergens treatments depends on environment factors (Tapia et al., 1997, Fayolle, 1996) and the sensitivity of hybrids (Bónis et al., 2000, Hart and Wax 1999). During the last years the number of corn and silage maize recognized by the state is largely improved, which is near to 400 together. These different genotypes significantly differ in their agrotechnological response, thus show a different hybrid reaction. It is also important to emphasize that all herbicides treatments mean a lower-higher stress effect on the growth-improvement of corn, which appears in the agronomical, fenometrical characteristics and also in the yield.

Materials and Methods

The small, plot experiments have been performed at the Experimental Station at Látókép of the University of Debrecen, Centre of Agriculture, Faculty of Agriculture, Department of Crop Production and Applied Ecology on calcareous chernozem soil. The soil of the experiment was nearly neutral (pH_{KCl}6,46), with average phosphorus and potassium content – according to soil examination - (Al soluble P_2O_5 133 mg/kg, Al soluble, K_2O 240 mg/kg). The soil of the experiment has favourable water husbandry and water holding characteristics, is in good condition, and perfectly suitable for corn production.
The force crop was winter wheat. The agrotechnical elements met the requirements of modern crop production.

In the experiment the following hybrids have been examined:

A De 377 SC
B Katinka
C Veronika
D Borbála
E Gazda
F Maraton
G Norma

The sowing of the experiment performed 22, April 2002 with 68.000 seeds/ hectare.
In the experiment the following herbicide-treatment have been adjusted:

1. Weedy control
2. Hoed control
3. Escort 4,0 l/ha (early post)
4. Merlin SC 0,22 L/ha + Dezormon 1,0 l/ha (early post)
5. Escort 4,0 l/ha (normal post)
6. Merlin SC 0,22 L/ha + Dezormon 1,0 l/ha (normal post)
7. Motivel 1,0 l/ha + Cambio 3,0 l/ha (normal post)
8. Titus 25 DF 40 g/ha + Callisto 0,25 l/ha + Trend 0,1% (normal post)
9. Motivel 1,0 l/ha + Cambio 3,0 l/ha (late post)
10. Titus 25 DF 40 g/ha + Callisto 0,25 l/ha + Trend 0,1% (late post)

The experimental treatments have been performed at the following time and state of development:

Hoeing (hand)	24, May 2002
	02, June 2002
Early postemergens	07, May 2002
	2-3 leaves of development
Normal postemergens	14, May 2002
	5 leaves of development
Late postemergens	20, May 2002
	7-8 leaves of development

The harvest of the experiment has been performed at 9, October 2002 with a Sampo combine.
In the range of the research project examinations of crop-dynamical, agronomical, plant-health, weed-dynamical, fitotoxical, crop producing factors have been accomplished, the yields and the water-content of seeds at harvesting have been measured.

Results and Discussion

The results of the experiments in 2002 prove that during the vegetation period the weed-coverage indicators – depending on the herbicide-treatments – increased form step by step. At the end of the vegetation period (1st treatment) the weed-coverage indicators of the weedy control were between 7,6% and 8,6%, while in the hoed control they were 4,3-5,1%. As a result of the herbicide-treatments the weed-coverage significantly decreased (measurements at 14, May; 20, May; 02, June).
The herbicide-treatments were characterized with adequate efficiency in this year's experiments, which have been proved by the weed-examinations before harvesting (time of measurement: 29, September):

3. treatment 4,9-6,0% weed-coverage
4. treatment 1,4-1,9% weed-coverage
6. treatment 1,4-1,7% weed-coverage
7. treatment 1,1-1,4% weed-coverage
8. treatment 1,0-1,4% weed-coverage
9. treatment 2,2-2,4% weed-coverage
10. treatment 2,1-2,4% weed-coverage

Adequate yields have been achieved as a result of favourable weather in 2002. The yields (without the extreme herbicide-treatment) were between 8-13 t ha^{-1} –according to the genotype, the time of herbicide-treatment and herbicide agent. In the vegetation period in 2002 the most favourable yields

(Table 1) were given by Maraton, Veronika and Norma hybrids (yields of 10-13 t ha^{-1}).

Table 1. Effects of herbicides on the yield of maize (average of hybrides, Debrecen, 2002)

Treatment		Yield kg/ha
1.	Weed control	9 875
2.	Hoed control	10 739
3.	Escort (early)	7 225
4.	Merlin+Dezormon (early)	9 331
5.	Escort (normal)	-
6.	Merlin+Dezormon (normal)	10 266
7.	Motivel+Cambio (normal)	10 016
8.	Titus+Callisto+Trend (normal)	10 374
9.	Motivel+Cambio (late)	9 552
10.	Titus+Callisto+Trend (late)	9 043

In the average of hybrids, we achieved similar yields to the hoed control (10.739 kg/ha) in the 6. treatment (Merlin+Dezormon normal post, 10.26 kg/ha), 7. and 8. treatments (Titus+Cambio+Trend normal post, 10.374 kg/ha) (Table 2.).

Table 2. Effects of herbicides on the yields of maize genotypes (Debrecen, 2002)

Hybrid	Weed control kg/ha	Hoed Control kg/ha	The best herbicide kg/ha	Yield difference (kg/ha)	
				To the weed control	To the hoed control
De 377 Sc	8 807	10 258	10 433	1 451	175
Katinka	8 990	10 247	9 764	1 257	-483
Veronika	10 455	11 094	10 893	639	-201
Borbála	9 449	9 860	9 429	411	-431
Gazda	9 447	10 514	10 263	1 067	-251
Maraton	12 531	13 022	12 693	491	-329
Norma	9 445	10 178	10 136	733	-42

The use of herbicides meant a lower-higher stress factor to the corn crops in all treatments, and it depends mostly on the time of use and the herbicide agent (Table 3).

Table 3.General and specific herbicide effects on the yield of difference maize genotypes (Debrecen, 2002)

Hybrid	Average of herbicides kg/ha	The best herbicide treatment kg/ha	Worst herbicide treatment kg/ha
De 377 Sc	9 471	10 433	8 453
Katinka	8 867	9 764	6 509
Veronika	10 111	10 893	9 247
Borbála	7 632	9 429	2 838
Gazda	7 812	10 263	7 213
Maraton	11 334	12 693	10 152
Norma	8 973	10 136	3 997

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Summary

HYBRID-SPECIFIC WEED CONTROL IN MAIZE PRODUCTION

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The rate of weed – coverage is affected by the herbicide, the time of use, the state of weeds and the competence of corn – hybrids.
The postemergens herbicides have a stress effect on the corn features and productivity.
The effect of stress depends on:

- time of use
- active substance of herbicide
- genotype

Besides the „general" herbicide – tolerance of the hybrids we must take into consideration the „specific" tolerance.

MAPPING THE DISTRIBUTION OF PERENNIAL WEED SPECIES FOR PLANNING PRECISION WEED CONTROL

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Perennial weeds cause a special part of weed problems. The species with dominating vegetative reproduction like *Cirsium arvense* (Béres et al. 2000, Lehoczky et al. 2003) can be detected on the plots through several years.
Therefore surveying them bears a prognostic value and the data received can be used for more years. Perennial weeds often spread in patches and precision weed control has special importance in protection against them.
Precise surveys provide further possibilities in the field of weed-biology research as perennial weeds can attract attention not only because of occupation of a place and their features but because of their nutrition needs and uptake (Lehoczky 1994, 2000).
The Balázs-Ujvárosi method is used for nation-wide surveys and analysis at company level in Hungary in the last decades. The method is based on estimating the weed cover (Reisinger 2000, 2001). The method is applied with 6 hectare sampling density therefore it provides only monitoring-type data on the weed flora in the relevant territory. As a result of our investigations a method based on sample taking cannot produce significant data to determine the distribution of weed species within a plot exactly, even if it uses a relatively high sampling density with 2-5 sample areas ha^{-1} (Reisinger et al. 2003).
The GPS technology (Stafford et al. 1996) contributes to the exact survey of perennial weed patches. With the help of a GPS receiver the contours, the position and location of the patches can be traced and recorded. Weed patches are able to infest from some percentage of a field to 80% (Brown et al. 1990, Thompson et al. 1991, Johnson et al. 1995, Rew et al. 1996). Perennial weed species can easily be surveyed on cereal stubble, in root plants in the period after field emergence. In that respect methods using remote sensing (Campbell 1996) can give an indication for planning the survey or they can even replace it (Christensen et al. 1994, Johannsen et al. 1998, Lamb et al. 2000).
Mapping the distribution of perennial weeds within a plot can help to make site-specific application plans, which can result in a reduction of herbicide use by 80%.

Materials and Methods

We carried out our investigations on a 7.4 ha large part of a maize field on 4th June 2003 in Mosonmagyaróvár. Maize plants were 30-40 cm high and the crop was in a stage before closing the rows. Pre-emergent treatment of the field (acetochlor 1600 g ha^{-1} + AD-67 160 g ha^{-1} and atrazine 900 g ha^{-1}) provided good results, *Cirsium arvense* (L.) Scop. and *Lepidium draba* L. were to be detected at high density only on smaller patches, which were located at the edges of the plots.

Weed patches were located with a Trimle Pathfinder Power GPS receiver by 5-7 satellite sensing at the same time on average with the application of Omnistar signal-correction of submeter accuracy. Field walking happened alongside a 20-30 m wide strip of land by walking around the patches on the way. Weed patches were recorded as polygons by registering their position at every 10 seconds. Patches smaller than $1m^2$ were not taken into account. We could find *Cirsium* patches easily as the infestation concentrated on patches with higher N-supply because of the uneven N-supply.

The data-file set up during the field-work was converted into an ESRI shapefile and was processed and maps were made using ArcView GIS 3.2 software. Our field walking revealed that the same weed species were surveyed repeatedly or overlapped by other patches in 14 cases so we united these patches on the map.

Overlapping of the contours of the plot was eliminated in order to circle the area with one line. The contours of the weed patches were corrected if necessary (overlapping, multi-polygons, not completely closed forms).

Results

With the help of the ArcView software and after the needed corrections of the data maps were produced, where the distribution of *Cirsium arvense* and *Lepidium draba* were described. 116 patches of *Cirsium arvense* and 4 patches of *Lepidium draba* were surveyed altogether in the field. After eliminating the repeated surveys and overlapping 106 patches of *Cirsium arvense* and 3 patches of *Lepidium draba* remained on the plot (Figures 1-3). Altogether 109 patches were surveyed on an area of 7.4 hectares.

Figure 1. *Lepidium draba* patches in the field

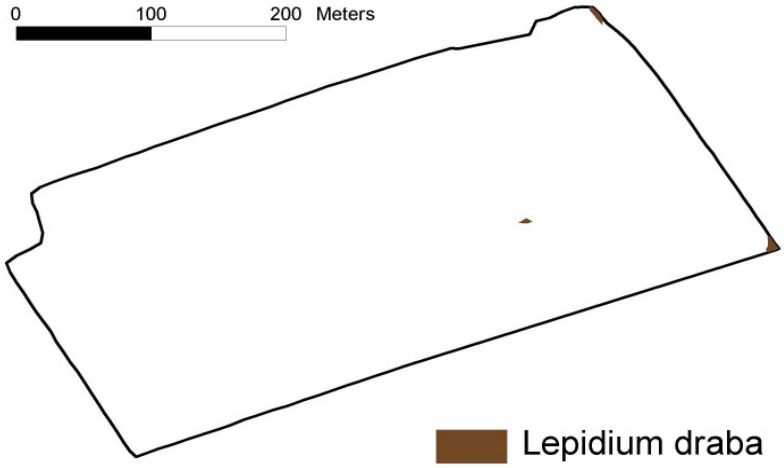

Figure 2. Distribution of *Cirsium arvense* in the field

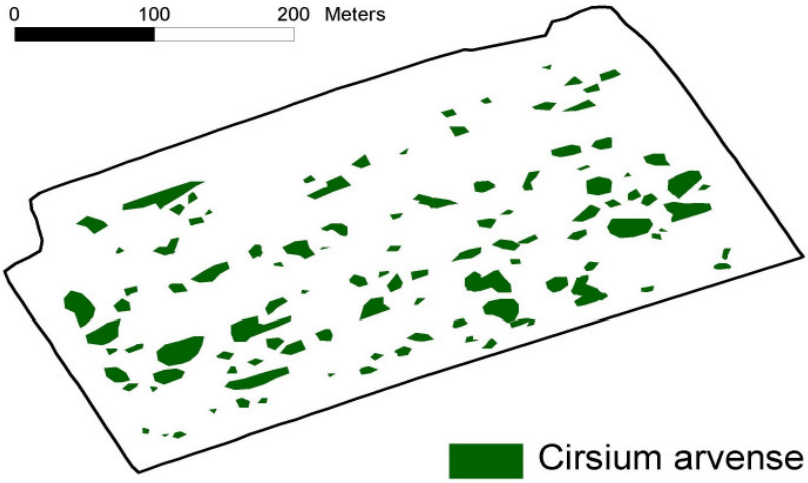

Figure 3. Distribution of *Cirsium arvense* and *Lepidium draba* in the field

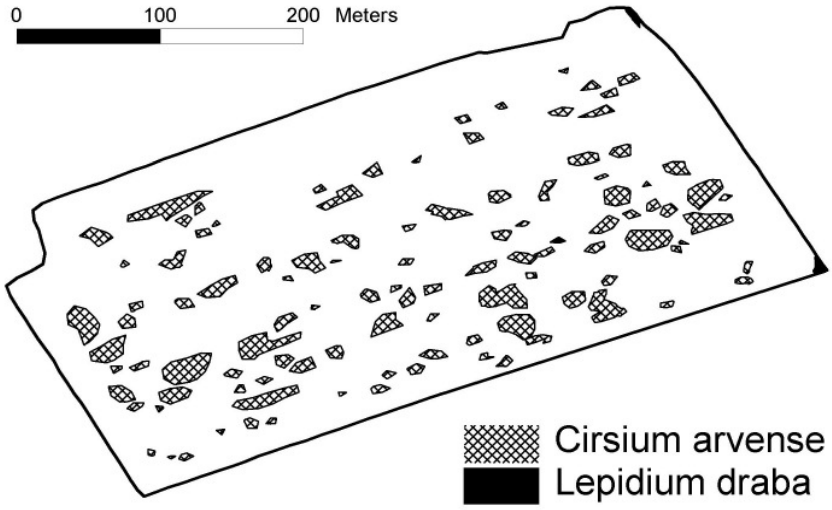

Table 1. Distribution of weed patches as per size, total and average area and the rate of area compared to the total area of the field

Area (m^2)	***Cirsium arvense*** **patch /pc**	***Lepidium draba*** **Patch /pc**
< 20	27	1
20-50	37	2
50-100	22	-
100-200	9	-
200-300	10	-
300 <	1	-
Totally:	106	3
Total area	7 067,59	89,79
Average	66,68	29,93
Area of the field	73 774	73 774
Rate of area(%):	9,58	0,12

The total area of *Cirsium arvense* covered 7 068 m^2 and *Lepidium draba* patches covered 90 m^2 altogether.

Since the total area of the field covers 73 774 m^2 they equal 9,6 respectively 12% of the total area. Therefore we can say that in spite of the great numbers of weed patches their total occurrence does not approach 10 % of the field area.
Analysing the data in Table 1 we can conclude that the average *Cirsium* patch size is 66,68 m^2, and *Lepidium* patches cover 29,93 m^2. Figures clearly show that the extension of weed patches is not strip-like, so the average diameter of a weed patch does not exceed 10 meters. As a result we recommend to section the spraying frame in order to carry out a precise application with maximum possible reduction of herbicide use on the test plot.
Investigating the distribution of the size of weed patches we can see that most of *Cirsium* patches are smaller than 100 m^2 and only one patch is larger than 300 m^2 (330 m^2). Most patches belong to the category of 20-50 m^2. *Lepidium* patches are smaller than 50 m^2.

Discussion

Patches of weed species on a plot can be detected with the help of a GPS receiver very accurately and efficiently. The spread of these weed species cannot accurately be mapped with the use of methods that are based on marking the sample area randomly or systematicly.
The performance of the survey depends on the number and size of weed patches, since patches fewer in number and larger at size can be registered more efficiently. According to our experience 2-5 ha $hour^{-1}$ can be surveyed by walking around the weed patches.
The efficiency of the method is greatly influenced by the smallest patch size that is still to be surveyed. Small size patches (much smaller than $1m^2$) cannot always be surveyed economically and stressing the zero tolerance contradicts the principle of weed control.
The smallest weed patch that is to be surveyed usually depends on the average patch size, the accuracy of the GPS receiver and the technical parameters of the automatic spraying equipment used in the application.
Larger areas can be surveyed more efficiently if we use a jeep or eventually a motorbike (quad).
If the soil is wet and the crop is low weed patches can be detected more easily and of course our own tracks, too. This fact can reduce overlapping and repeated surveys. Also, if we mark the surveyed patches it will help to avoid repeated detection.
If an air-photo of the area infested with perennial weeds is available it can be used to plan the work. If there is a chance to make an ortho-image from this air-photo a field walking can be avoided.

Walking around the weed patches is relatively subjective especially if the population density is low. It is obvious that dense weed populations should be surveyed.

The method is of special importance in root crop growing when there are only perennial weeds in the area after an efficient basic treatment. We can save even 70-80% of the herbicide quantity if we carry out a site-specific treatment based on the data of the survey. Walking around the weed patches registered by GPS perennial every weed species can be registered accurately, but this method can also be used for detecting annual weed species occurring in patches owing to certain causes (low lying field parts, altering soil patches etc.).

With the help of the weed patch maps supplemented by the necessary puffer areas and adequate software we can prepare plan of herbicide application for the automatic sprayers guided by GPS. The rate of the area was infested by perennial weed species do not reach 10% of the total area involved into our investigations.

If we survey the perennial weed patches for several years it will provide us information about the changes in population dynamic.

If we survey perennial weed species once it can be provide information for several years due to the reproduction practice.

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Summary

MAPPING THE DISTRIBUTION OF PERENNIAL WEED SPECIES FOR PLANNING PRECISION WEED CONTROL

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Protection against perennial weed species is a special field of weed control. Postemergent technologies can be applied against these species effectively, either alone or after pre-emergent treatments. Perennial weed species often spread on farm lands in patches. Site-specific treatment of the patches help to save a considerable amount of herbicides. The aim of our trial is to survey the distribution of the patches of *Cirsium arvense* (L.) Scop. and *Lepidium draba* L. in maize by DGPS as well as to analyse the rate of the patch size compared to the territory and followed by making maps for planning precision treatments.

CROP-WEED COMPETITION: *CANNABIS SATIVA* L. IN WINTER WHEAT

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Industrial hemp (*Cannabis sativa* L.) has been grown for its long and high-quality fibers for many centuries. However, its importance has been decreased during the last decade by the introduction of synthetic fibers, the mechanization of cotton production, and drug prohibition laws [1,2]. Industrial hemp is still grown in Hungary, but, since its acreage has been reduced to less than 5000 ha, it is not listed any more among the important crop plants of the country [3]. However, recently a wild variety (*Cannabis sativa* ssp. *spontanea*) of industrial hemp has appeared as a fast-growing, tall weed in almost all of the agricultural lands in Hungary. The plant forms sizeable patches in cereals, especially in areas where the density of the crop plant is reduced by poor germination, plant disease or low crop vigor [3]. Wild hemp became a problematic weed, because a) it reduces the crop yield *via* competition for light, water, and nutrients, b) it also contains a number of biologically active (antimicrobial, allelopathic, and narcotic) substances [4], and c) it is a host of the parasitic plants *Orobanche* and *Cuscuta* spp. [5].

The introduction of information technology resulted in major advances in site- and time-specific crop management. Thus, mapping of fields for differences in nutrient concentrations, pH, and physical properties has allowed the application of fertilizers in different doses at selected parts of the field, resulting in ecological and financial benefits [6]. However, in case of weed control, uncertain knowledge on weed seed distribution, viability, and short and long term responses to control measures make site- and time-specific technologies less than obvious. Therefore, on-farm research is necessary to assess all the uncertainties related to weed management [6,7]. The objectives of this research were to develop a site-specific wild hemp management method in winter wheat based on the competitiveness of this weed plant.

This study was initiated to determine the competitiveness of wild hemp against winter wheat and other weeds in order to develop a wild hemp

management method. Therefore, we measured the dominance properties and the biomass production of wild hemp in a winter wheat field and compared them with those of the crop plant. The literature regarding fertilization requirements for industrial hemp consistently indicates a need for NPK application (generally at a rate consistent with wheat production), because hemp removes large quantities of minerals from the soil [1,8]. Therefore, nutrient uptake by wild hemp and winter wheat were also determined and compared.

Materials and Methods

The experiments were carried out on April 3, 2002 in a 36-hectare wheat field at Baracska (Fejer county, Hungary; soil properties are listed in Table 1). Wheat (cv Gyozo, Martonvasar Seeds, Martonvasar, Hungary) was sown on October 16, 2001, following the application of NPK (nitrogen, phosphorus, and potassium) fertilizer (39, 78, and 78 kg ha^{-1}, respectively). On March 16, 2002, a further 50 kg ha^{-1} nitrogen fertilizer was applied. Crop was harvested on July 10, 2002 (average yield was 4.46 t ha^{-1}).

Table 1. Soil properties (means [variances]) of the project area (Baracska, Fejer county, Hungary)

Type		**Texture**	**Organic matter**, %	**pH**	**$CaCO_3$** %	**P_2O_5** mg kg^{-1}	**K_2O** mg kg^{-1}
FAO	USDA						
Calcaric phaeosem	Mollisol	Loam	3.2 [0.83]	7.5 [1.8]	5.7 [1.2]	283 [54]	314 [39]

For evaluating weed populations the 36-hectare project area was divided to 18 x 250 m blocks (altogether 80, 0.45 ha each) and within the blocks 2 x 2 m sampling areas were assigned and identified by GPS coordinates. Weed populations were assayed by using the Balázs-Ujvárosi coenological method [5], and samples of crop (wheat) and weed plants were taken and analyzed for phosphorus, potassium, nitrogen and calcium concentrations by using spectrophotometric and flame photometric methods [9,10]. From the recorded plant population densities dominance values of the weed species were determined and weed maps were constructed by using Imagine 8.5 Professional software (Erdas, Atlanta, GA, USA).

Results and Discussion

Although wild hemp has been considered a late summer weed in Hungary [5], our studies indicate that it can massively germinate early in the spring even in established and well growing wheat. At the time of weed survey (April 3, 2001) high numbers of 2-4 leaf stage wild hemp plants were observed (Figure 1) and wild hemp had the highest coverage among the 15 weed species identified in the field (Table 2). Wild hemp plant numbers reached 302 plants m^{-2} (Figure 1) (average 143 plants m^{-2}), and coverage values as high as 25.8 % (average 11.6 %) were recorded. Still, in spite of its large plant density at the time of our weed survey the net biomass (on dry weight basis) of wild hemp was only 1.1 % of the crop plant (Table 3).

Figure 1. Wild hemp (Cannabis sativa spp. spontanea) weed map of the project area (April 4, 2002; Baracska, Fejer county, Hungary) with 5% weed cover interval contour lines. Lowest and highest weed densities: A-weed cover 0%; B-weed cover >25% to 30%

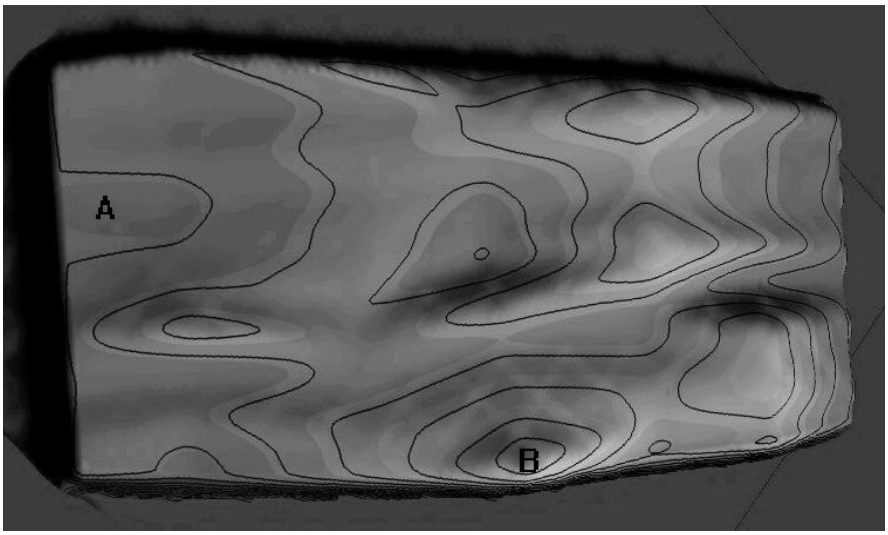

Table 2. Coverage of weeds in the project area (Baracska, Fejér County, Hungary)

Rank	**Weed species**	**Coverage**, %
1.	*Cannabis sativa* ssp. *spontanea*	11.61
2.	*Sisymbrium sophia*	0.28
3.	*Papaver rhoeas*	0.25
4.	*Bilderdykia convolvulus*	0.20
5.	*Cirsium arvense*	0.14
6.	*Chenopodium album*	0.04
7.	*Chenopodium hybridum*	0.02
8-15.	*Others*	0.04
	Total weeds	12.57

A comparison of the nutrient content of the aerial plant parts showed that winter wheat contained significantly higher concentration of nitrogen than wild hemp (Table 3).

Table 3. Nutrient element content (expressed in percentage of dry weight) and biomass (fresh and dry weight) of winter wheat (*Triticum aestivum* L.) and wild hemp (*C. sativa* ssp. *spontanea*), (Baracska, Fejér County, Hungary)

Nutrients in plants (dry weight%) **and plant biomass (**g m^{-2}**)**		**Winter wheat**	**Wild hemp**	**$LSD_{5\%}$**
Nitrogen		3.66 ± 0.48	2.52 ± 0.26	0.29
Phosphorus		0.37 ± 0.04	0.48 ± 0.05	0.04
Potassium		3.01 ± 0.32	2.86 ± 0.37	0.26
Calcium		0.54 ± 0.04	2.17 ± 0.45	0.24
Biomass	Fresh weight	1607 ± 374	24.0 ± 13.9	201
	Dry weight	532 ± 105	5.95 ± 0.40	56

However, other data in Table 3 clearly indicate that wild hemp competes with winter wheat for water and for all of the mineral nutrients studied (especially for phosphorus and calcium). The high competitiveness of wild hemp for nutrients compares well with the high nutrient needs of industrial

hemp varieties [1,2,8]. Our data also show that early in the vegetation period nutrient uptake by wild hemp plants growing in winter wheat crop is relatively small. However, with the rise of the daily average temperatures wild hemp grows more rapidly and soon reaches the height of the wheat plants (data not shown). Therefore, to keep a low competitiveness of the weed, a postemergent herbicide treatment to suppress wild hemp is necessary. Thus, after localization the patches of wild hemp and other weeds in the field, weed contour maps were created, divided into manageable blocks, and a site-specific weed control technology was designed.

Acknowledgements

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Summary

CROP-WEED COMPETITION: *CANNABIS SATIVA* L. IN WINTER WHEAT

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Wild hemp (*Cannabis sativa* ssp. *spontanea*) is a rapidly spreading weed in Hungary that forms large and stable patches in most agricultural fields. To evaluate the competitiveness and the nutrient uptake of wild hemp experiments were carried out in a 36-hectare agricultural field, using 80 evenly distributed sampling areas that were identified by GPS coordinates. Field maps of wild hemp infestations were formulated to determine the density and dominance of this weed, and plants were analyzed for macroelement (nitrogen, phosphorus, potassium, and calcium) contents.

Wild hemp showed significant competitiveness against winter wheat, greatly reducing the availability of nutrients to the crop plant. After localizing patches of wild hemp and other weeds in the field by GPS coordinates a weed contour map was created. These maps were divided into manageable blocks and a site-specific weed control technology was designed in order reduce herbicide use, thereby decreasing the costs and environmental impact of wild hemp control.

SPREAD OF ALLELOPATHIC WEEDS IN CULTIVATED AREAS IN HUNGARY, THE ROLE OF ALLELOPATHY IN SPREADING

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Professionals have been monitoring weed infestation, spreading and abundance of the particular weed plants in Hungarian cultivated areas since 1950. Four national surveys have been conducted to the present (1950, 1970, 1988 and 1997). In addition to their botanical importance, such surveys have been of great interest for plant protection, because one of the most important elements of successful weed control is to know: which species are present at a particular place and how high their abundance maybe. Moreover, weed management programmes cannot be developed without being aware of the trends of weed spreading.

As a result of the surveys we could monitor changes of weed populations on cultivated areas (Table 1).

Table 2 shows the weed species whose increased spreading and infestation area have been conspicuous in the past 50 years.

We wanted to find the ecological and economic reasons for increased or decreased importance of certain weeds. During the in-depth studies, in the cases of the species with especially high increase, we found several relationships, such as changes of crop management programmes, building-up of resistance, slow warming and the use of new herbicides on extended areas. Studying these causes for *Ambrosia artemisifolia, Cirsium arvense, Abutilon theophrasti, Asclepias syriaca*, dividing them to factors under the influence of men and to ones independent of human activity, we found the following share given in Annexes 1-4.

Table 1. Monitoring of weed population changes in field areas

	*	**	Rank	Cover %	Rank	Cover %	Rank	Cover %	Rank	Cover %
AMBROSIA ARTEMISIIFOLIA L.	R	A	21	0,3926	8	0,8734	4	2,5724	1	4,7030
ECHINOCHLOA CRUS-GALLI P.B. VAR.	R	-	9	0,8557	1	3,7280	1	4,4192	2	3,9095
AMARANTHUS RETROFLEXUS L.	R	A	17	0,5079	5	1,4658	3	3,0610	3	3,6290
CHENOPODIUM ALBUM L.	R	A	3	1,5319	3	2,0662	2	3,0816	4	2,8988
CIRSIUM ARVENSE (L.) SCOP.	R	A	2	2,0031	7	1,1245	8	0,7090	5	1,8070
MATRICARIA INODORA L.	R	A	66	0,0657	26	0,2316	6	1,2984	6	1,5429
CONVOLVULUS ARVENSIS L.	R	-	1	7,9266	2	2,5144	5	1,9439	7	1,4532
DATURA STRAMONIUM L.	R	A	177	0,0055	59	0,0619	19	0,3847	8	1,0694
AMARANTHUS CHLOROSTACHYS WILLD.	R	-	105	0,0231	18	0,3948	13	0,5691	9	0,9435
GALIUM APARINE L.	R	-	137	0,0103	50	0,0875	12	0,5858	10	0,8716
SORGHUM HALEPENSE (L.) PERS.	R	A			94	0,0249	18	0,4040	11	0,8204
ELYMUS REPENS (L.) GOULD	-	A	27	0,2800	12	0,5065	20	0,3845	12	0,6483
PANICUM MILIACEUM L.	-	-	199	0,0032	192	0,0045	23	0,2905	13	0,6027
XANTHIUM STRUMARIUM L. SSP. STUM.	R	A	130	0,0129	113	0,0148	24	0,2709	14	0,5752
POLYGONUM LAPATHIFOLIUM L.	R	-	29	0,2524	16	0,3994	10	0,6060	15	0,5273
BILDERDYKIA CONVOLVULUS L.	R	-	14	0,7110	6	1,1441	11	0,600	16	0,5210
APERA SPICA-VENTI (L.) BEAUV.	R	-	56	0,0762	36	0,1435	14	0,4617	17	0,4896
HELIANTHUS ANNUUS L.	R	A	206	0,0030	119	0,0141	16	0,4245	18	0,4892
SETARIA GLAUCA (L.) P.BEAUV.	R	-	7	1,1054	4	1,9544	7	0,7208	19	0,4872
PAPAVER RHOEAS L.	R	-	24	0,3505	21	0,3193	15	0,4293	20	0,4664

Table 2. Changes in importance of weed species between 1950 and 1977

Practically important weed species of maize on the base of 1977 year data

Species with prominent importance		1950		1970		1988		1997	
		Rank	Cover%	Rank	Cover%	Rank	Cover%	Rank	Cover%
ECHINOCHLOA CRUS-GALLI P.B. VAR.	W	2	2,0247	1	6,4906	1	7,4392	1	6,4575
AMARANTHUS RETROFLEXUS L.	H	8	0,8413	4	2,6643	2	4,8401	2	6,3022
AMBROSIA ELATIOR L.	H	20	0,4116	8	0,9556	4	3,3169	3	5,9505
CHENOPODIUM ALBUM L.	H	4	1,8853	2	3,0088	3	4,3444	4	3,9594
CONVOLVULUS ARVENSIS L.	W	1	7,6541	3	2,8449	5	2,4996	5	1,7155
DATURA STRAMONIUM L.	W	101	0,0101	34	0,1274	12	0,6171	6	1,6534
CIRSIUM ARVENSE (L.) SCOP.	H	3	1,9751	6	1,1096	10	0,7435	7	1,6220
AMARANTHUS CHLOROSTACHYS WILLD.	H	69	0,2441	17	0,4601	8	0,8672	8	1,4805
SORGHUM HALEPENSE (L.) PERS.	W			57	0,2994	14	0,5690	9	1,2292

Table 2 (continued)

Species with importance		1950		1970		1988		1997	
		Rank	Cover%	Rank	Cover%	Rank	Cover%	Rank	Cover%
PANICUM MILIACEUM L.	-	40	0,0907	137	0,0039	17	0,4818	10	0,9883
ELYMUS REPENS (L.) GOULD	-	18	0,4723	12	0,6419	16	0,4866	11	0,8570
XANTHIUM STRUMARIUM L. SSP. STUM.	W	71	0,0229	71	0,0176	18	0,4328	12	0,7712
POLYGONUM LAPATHIFOLIUM L.	W	23	0,3479	13	0,5137	9	0,8660	13	0,7639
SETARIA GLAUCA (L.) P.BEAUV.	W	6	1,4587	5	2,3588	6	1,1061	14	0,7207
HIBISCUS TRIONUM L.	-	13	0,6605	9	0,8089	11	0,6799	15	0,6170
SINAPIS ARVENSIS L.	W	15	0,6578	7	0,9645	7	0,9548	16	0,5389
HELIANTHUS ANNUUS L.	W			124	0,0054	22	0,3001	17	0,4446
ABUTILON THEOPHRASTI MEDIC.	W	220	0,0005			43	0,0755	18	0,4236
CHENOPODIUM HYBRIDUM L.	-	43	0,0712	35	0,1593	24	0,2523	19	0,4179
AMARANTHUS BLITOIDES S.WATS.	H	63	0,0302	31	0,1600	20	0,3231	20	0,4027

Looking for the common factor or trait the drastic spread can be attributed to, considering a part of the rapidly increasing weeds in Hungary, *Ambrosia artemisifolia, Amaranthus retroflexus, Chenopodium album, Cirsium arvense, Matricaria inodora, Datura stramonium, Sorghum halepense, Elymus repens, Helianthus annuus, Xanthium spp., Abutilon theophrasti, Asclepias syriaca*, we found only one factor out of human control for the time being, which can be observed in all of the listed weed species, namely **ALLELOPATHY**. We concluded that the weed species possessing this trait has an advantage under Hungarian conditions over the majority of the other weed species and the crops during their competition. Their abundance is increasing accordingly, and they also spread more rapidly than the majority of the other occurring weeds. Photos (1-6) show how highly the populations of the above species can increase.

ANNEX 1

MAIN REASONS OF MASS POPULATION INCREASE OF COMMON RAGWEED *(AMBROSIA ARTEMISIFOLIA)* INFLUENCED BY HUMAN ACTIVITY

- HOBBY GARDENS
- USE OF SEEDS INFESTED WITH WEED SEEDS
- HUMAN FACTOR (RAGWEED IS A NICE PLANT)
- SHORTAGE AND HIGH COST OF SUCCESSFUL WEED MANAGEMENT PROGRAMMES
- CONTRACTED WORK (IMPLEMENTS FOR CULTIVATION, HARVESTING, TRANSPORT)
- RESISTANCE
- INCREASE OF FALLOW AREAS
- STUBBLES LET UNCULTIVATED
- MISSING OF INITIAL CONTROL (WHEN ONLY SOME PLANTS OF THE WEED ARE PRESENT IN THE FIELD)

- LACK OF PROFESSIONAL SKILLS (SMALL-SCALE FARMERS)
- SHORT-TERM EFFECT OF UREA HERBICIDES
- INCREASE OF WEEDY FIELD EDGES
- LOW FREQUENCY OF CROP ROTATION PRACTICE (BI-AND MONOCULTURE)
- BIG CONSTRUCTIONS LASTING FOR SEVERAL YEARS (e.g. HIGHWAYS)

REASONS OF MASS POPULATION INCREASE OF COMMON RAGWEED *(AMBROSIA ARTEMISIFOLIA)* OUT OF HUMAN CONTROL

- CHANGED BIOLOGY OF GERMINATION (GERMINATION ABILITY REMAINS FOR 15-20 YEARS)
- ALLELOPATHY
- LACK OF NATURAL ENEMIES
- INCREASED COLD HARDINESS
- GOOD COMPETITION ABILITY (E.G. IN SUNFLOWER RAGWEED CAN REACH 3-3,5 M HEIGHT)
- RESISTANCE TO DRAUGHT
- POOR HERBICIDE CHOICE FOR EXTREME SOILS (e.g. SAND)
- HIGH REGROWTH ABILITY
- SOIL TYPES
- DECREASING COMPETITIVE ABILITY OF CROPS
- INCREASE OF FROST-FREE DAYS IN THE PAST 10-15 YEARS
- LONG GERMINATION PERIOD (FROM APRIL TO SEPTEMBER)
- POOR PALATABILITY FOR VERTEBRATES

ANNEX 2

MAIN REASONS OF MASS POPULATION INCREASE OF CANADA THISTLE (*CIRSIUM ARVENSE*) INFLUENCED BY HUMAN ACTIVITY

- SHALLOW TILLAGE
- STUBBLES LET UNCULTIVATED FOR A LONG TIME
- INCREASE OF FALLOW AREAS
- SPLITTING OF FIELDS
- REDUCED AREAS WITH APPLICATION OF HERBICIDES WITH HORMONAL ACTIVITY
- REDUCED CEREAL AREAS WITH WEED CONTROL (ESPECIALLY SMALL-SCALE FARMERS)
- USE OF SEEDS INFESTED WITH WEED SEEDS
- WRONG CROP ROTATION PRACTICE (E.G. WHEAT - SUNFLOWER)
- SIGNIFICANT INCREASE OF SUNFLOWER AREAS
- USE OF POOR QUALITY SEEDS (SEEDS HARVESTED BY FARMERS)
- HIGH PRICE OF THE SELECTIVE HERBICIDE (LONTREL 300)
- LACK OF PROFESSIONAL SKILLS (SMALL-SCALE FARMERS)

- CONTRACTED WORK (IMPLEMENTS FOR CULTIVATION, HARVESTING, TRANSPORT)
- LOW DENSITY CEREAL STANDS (ITS MAIN REASONS)
- CHANGES IN CROP MANAGEMENT PROGRAMMES
— INAPPROPRIATE USE OF SULFONYLUREAS

REASONS OF MASS POPULATION INCREASE OF CANADA THISTLE (*CIRSIUM ARVENSE*) OUT OF HUMAN CONTROL

- MILD WINTERS
- RESISTANCE
- ALLELOPATHY
- DIFFERENT LEVELS OF RESISTANCE TO HERBICIDES OF THE FOUR TYPES OF THE WEED
- SOIL TYPE
- ANNUAL INCREASE OF FROST-FREE DAYS

ANNEX 3

MAIN REASONS OF MASS POPULATION INCREASE OF VELVETLEAF *(ABUTILON THEOPHRASTI)* INFLUENCED BY HUMAN ACTIVITY

- USE OF SEEDS INFESTED WITH WEED SEEDS
- NEGLECTED DITCHES AND WATER CHANNELS
- ORGANIC MANURE (WEED SEEDS GET THROUGH THE INTESTINAL TRACT OF ANIMALS UNAFFECTED)
- INAPPROPRIATE CROP ROTATION (SUGARBEET – SUNFLOWER)
- OMISSION OF STUBBLE STRIPPING
- CONTRACTED WORK (IMPLEMENTS FOR CULTIVATION, HARVESTING, TRANSPORT)
- LACK OF PROFESSIONAL SKILLS (POOR KNOWLEDGE OF WEEDS AND HERBICIDES

REASONS OF MASS POPULATION INCREASE OF VELVETLEAF *(ABUTILON THEOPHRASTI)* OUT OF HUMAN CONTROL

- ANNUAL INCREASE OF FROST-FREE DAYS (VELVETLEAF IS A PLANT OF HIGHER TEMPERATURE DEMAND)
- ALLELOPATHY
- ELAYED EMERGENCE
- RESISTANCE
- SOIL TYPE

ANNEX 4

MAIN REASONS OF MASS POPULATION INCREASE OF COMMON MILKWEED (*ASCLEPIAS SYRIACA*) INFLUENCED BY HUMAN ACTIVITY

- INCREASE OF FALLOW AREAS
- SHORTAGE OF WEED CONTROL PROGRAMMES AND EFFICIENT HERBICIDES
- STUBBLES LET UNCULTIVATED FOR A LONG TIME
- HUMAN FACTOR (RAGWEED IS A NICE PLANT, IT IS EVEN PROPAGATED)
- OMISSION OF STUBBLE TREATMENTS
- CONTRACTED WORK (IMPLEMENTS FOR CULTIVATION, HARVESTING, TRANSPORT)
- INCREASE OF WEEDY FIELD EDGES

REASONS OF MASS POPULATION INCREASE OF COMMON MILKWEED (*ASCLEPIAS SYRIACA*) OUT OF HUMAN CONTROL

- ANNUAL INCREASE OF FROST-FREE DAYS (COMMON MILKWEED IS A PLANT OF HIGHER TEMPERATURE DEMAND)
- DRY WEATHER PERIODS
- ALLELOPATHY
- INCREASED REGROWTH ABILITY
- SOIL TYPE

ACTUALITIES IN THE PLANT PROTECTION OF PROTECTED VEGETABLE CROPS
(Summary)

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In the southern regions of Hungary, large acreages of vegetables grown under glasshouses and plastics, are located. Protected crops are the main source of employment for several thousands of families in Hungary. Protected pepper (*Capsicum annum* L.) is grown on about 2,250 hectares (ha) while protected tomato (*Lycopersicon esculenum* L.) is grown on about 1,200 ha (1999 data). This represents about 64% of the total greenhouse area of 5400 ha in Hungary.

Under the conditions found in the South-Eastern part of Hungary, and particularly in sandy soils, *Meloidogyne* spp. (mainly *M. hapla* and *M. incognita*) nematodes are the most important pests. The high level of infestation causes significant losses , so soil application has to be performed on a routine basis.

In the last few years the cotton-bollworm (*Helicoverpa armigera)* became one of the most severe pests in greenhouse production, too. Its grub attacks mostly pepper and tomato. The damaged yield usually rots. The effective treatment is a difficult task, because the larva lives hidden. The foliage has to be covered permanently by insecticides.

The most severe diseases of the protected crops (pepper, tomato) are caused by viruses. The average infection level is generally about 10-60%, but in some cases 100% virus infection can be detected in pepper and tomato.

Regular epidemic survey has been made since the 1970's. Studies confirmed that the TMV (*Tobacco mosaic virus*), ToMV (*Tomato mosaic virus*), CMV (*Cucumber mosaic virus*), AMV (*Alfalfa mosaic virus*), and PVY (*Potato Y virus*) are the most wide-spread viruses causing severe yield losses, particularly if complex infection occurs. Since 1994, spread of TSWV (*Tomato spotted wilt virus*) has been observed, causing however, severe damages. During the past years, the spread of thrips virus vectors, namely *Frankliniella occidentalis* Pergande, has caused explosive appearance of TSWV. The spread of this virus has still not been general, actually it occurs in isolated locations particularly in the southern regions of Hungary. One can expect the spread of TSWV in the future, if no preventive measures are taken, the most important being the eradication of the possible virus sources, weed control and effective control of thrips.

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